High-Temperature Superconductivity Turns 20 >> page 1072

17 November 2006 | $10

Science

Materials Science

COMPOSITES

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SLIPPERY WHEN WET. Sample can easily be lost when tubing leaks or clamps slip off.	SAMPLE RECOVERY	>95% sample recovery.
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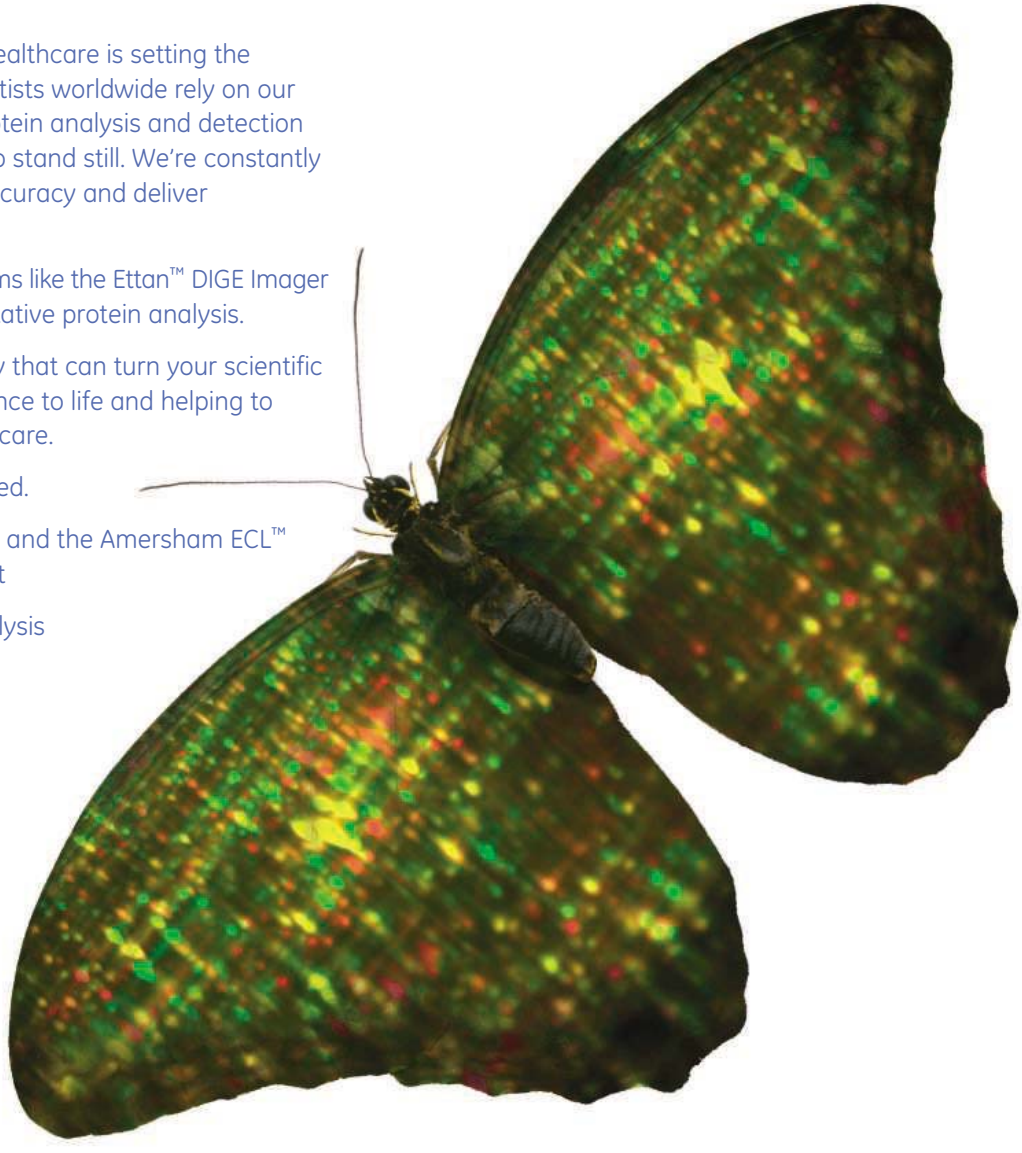

imagination at work

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Björkgatan 30, 751 84 Uppsala, Sweden.

GE08-06

Volume 314, Issue 5802

COVER

Fluorescence optical microscopy image of cadmium selenide nanoparticles. These polyethylene glycol–functionalized nanoparticles have segregated to cracks in a composite film confined between a brittle silicon oxide layer and a silicon substrate. Field of view is 30 by 50 μm. See the special section on materials science beginning on page 1099.

Image: S. Gupta and Q. Zhang

For related online content, see page 1043

DEPARTMENTS

1043 *Science* Online
1045 This Week in *Science*
1050 Editors' Choice
1054 Contact *Science*
1057 Random Samples
1059 Newsmakers
1161 New Products
1162 *Science* Careers

EDITORIAL

1049 Don't Grandfather Coal Plants
by M. Granger Morgan

SPECIAL SECTION

Materials Science: Composites

INTRODUCTION

The Right Combination 1099

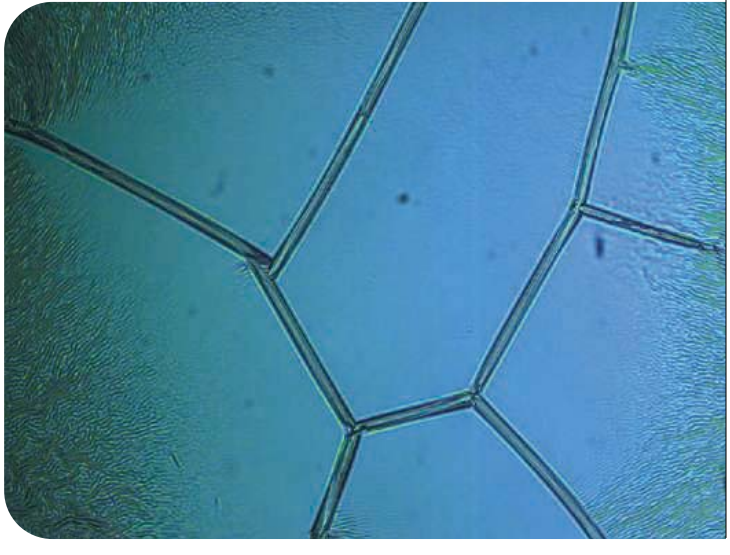
PERSPECTIVE

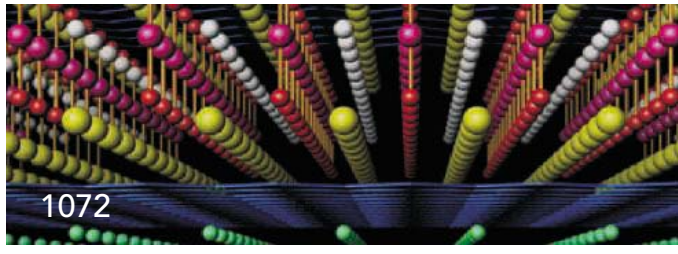
Composites in Armor 1100
P. J. Hogg

REVIEWS

In Quest of Virtual Tests for Structural Composites 1102
B. Cox and Q. Yang

Nanoparticle Polymer Composites: Where Two Small Worlds Meet 1107
A. C. Balazs, T. Emrick, T. P. Russell





NEWS OF THE WEEK

Election 2006

Science Awaits Impact of Democratic Sweep in Congress 1060
Environmentalists See a Greener Congress 1061
Gordon Steps Up to House Science Post 1061
Stem Cell Supporters Hail Results, But Political Lessons Aren't Clear 1062
Scientists Get Out the Word 1063
Elsewhere on the Election Front 1063
False Alarm: Atlantic Conveyor Belt Hasn't Slowed Down After All 1064
Novartis Invests $100 Million in Shanghai 1064
Electronic Nuisance Changes Its Ways 1065
>> *Report p. 1121*
SCIENCESCOPE 1065
SARS and Bird Flu Veteran to Take WHO Helm 1067

NEWS FOCUS

The Dawn of Stone Age Genomics 1068
>> *Research Article p. 1113*
A Neandertal Legacy?

Special Report: High-Temperature Superconductivity Turns 20

High T_c: The Mystery That Defies Solution 1072
>> Science *Express Reports by T. Valla* et al. *and K. Tanaka* et al.
The Next Big Hurdle: Economics 1075
Determined Duo Scored a Victory for Small-Scale Science 1078

CONTENTS continued **>>**

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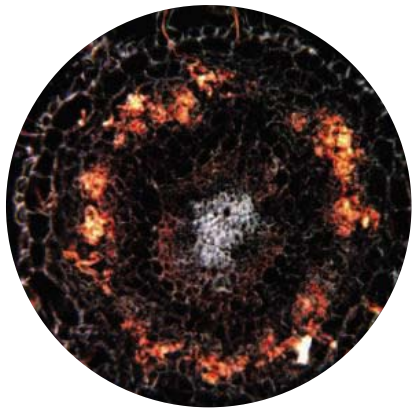

SCIENCE EXPRESS

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PHYSICS

The Ground State of the Pseudogap in Cuprate Superconductors
T. Valla, A. V. Fedorov, J. Lee, J. C. Davis, G. D. Gu
The existence of an energy gap in a nonsuperconducting cuprate suggests that a comparable gap in superconductors arises as electrons pair up but are not fully coherent.
>> News story p. 1072
10.1126/science.1134742

PHYSICS

Distinct Fermi-Momentum–Dependent Energy Gaps in Deeply Underdoped Bi2212
K. Tanaka et al.
Spectrometry on a high-temperature superconductor lacking a few of its electrons reveals that two additional energy gaps separate the pseudogap and the true superconducting gap.
>> News story p. 1072
10.1126/science.1133411

PLANT SCIENCE

A Cytokinin Perception Mutant Colonized by *Rhizobium* in the Absence of Nodule Organogenesis
J. D. Murray, B. J. Karas, S. Sato, S. Tabata, L. Amyot, K. Szczyglowski
10.1126/science.1132514

A Gain-of-Function Mutation in a Cytokinin Receptor Triggers Spontaneous Root Nodule Organogenesis
L. Tirichine et al.
In the legume Lotus, symbiotic, nitrogen-fixing bacteria induce formation of the root nodules in which they reside by eliciting a growth response from the plant itself.
10.1126/science.1132397

LETTERS

Why Aren't There More Scientists Advocating for Funding? *R. D. Wells and P. Farnham* 1081
Fighting Waterborne Infectious Diseases *R. C. Spear* et al. **Response** *A. Fenwick*
Debating the Worth of NCCAM Research *S. Folkman* et al. **Response** *D. M. Marcus and A. P. Grollman*

BOOKS *ET AL.*

The Regulatory Genome Gene Regulatory Networks in Development and Evolution *E. H. Davidson, reviewed by D. Arendt* 1085
The Reluctant Mr. Darwin An Intimate Portrait of Charles Darwin and the Making of His Theory of Evolution *D. Quammen, reviewed by J. Browne* 1086

POLICY FORUM

NIH in the Post-Doubling Era: Realities and Strategies *E. A. Zerhouni* 1088

PERSPECTIVES

Money Is Material *C. B. Burgoyne and S. E. G. Lea* >> Report p. 1154 1091
Why Do Freezing Rocks Break? *B. Hallet* >> Report p. 1127 1092
What Do Robots Dream Of? *C. Adami* >> Report p. 1118 1093
Sara Splits the Signal *J. A. Knoblich* >> Report p. 1135 1094
Breaking the H_2 Marriage and Reuniting the Couple *G. J. Kubas* >> Report p. 1124 1096
RNA Polymerase, a Scrunching Machine *J. W. Roberts* >> Reports pp. 1139 and 1144 1097

BREVIA

ECOLOGY

Rapid Temporal Reversal in Predator-Driven Natural Selection *J. B. Losos, T. W. Schoener, R. B. Langerhans, D. A. Spiller* 1111
As island lizards shift from ground to trees to escape predators, the selective pressure favors longer legs instead of the shorter legs favored on the ground.

RESEARCH ARTICLE

EVOLUTION

Sequencing and Analysis of Neanderthal Genomic DNA *J. P. Noonan* et al. 1113
The sequences of DNA fragments from Neanderthal bones date the divergence of humans and Neanderthals to about 370,000 years ago.
>> News story p. 1068

REPORTS

COMPUTER SCIENCE

Resilient Machines Through Continuous Self-Modeling *J. Bongard, V. Zykov, H. Lipson* 1118
When it receives appropriate sensory information, a mobile robot can compensate for damage to one of its four legs by updating an internal model of itself. *>> Perspective p. 1093*

PHYSICS

Solid-State Thermal Rectifier *C. W. Chang, D. Okawa, A. Majumdar, A. Zettl* 1121
Systematically increasing the amount of a platinum compound along the length of a boron or carbon nanotube allows heat to flow preferentially in the opposite direction.
>> News story p. 1065

CONTENTS continued **>>**

Shine Light on microRNA Expression Profiles

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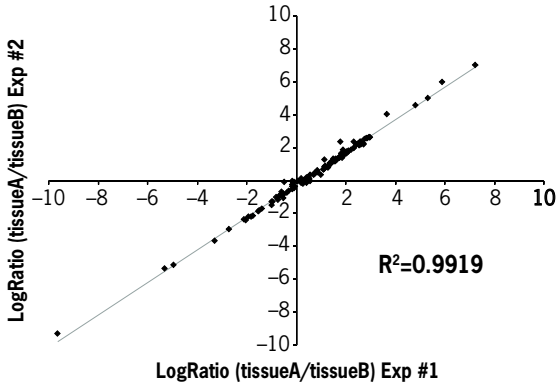

Reproducibility of Differential Expression Ratios with *mir*Vana™ miRNA Bioarrays. Relative miRNA expression levels in tissues A and B were determined by analysis of each tissue on two independent *mir*Vana™ miRNA Bioarrays and comparison of the resulting normalized signal intensities [$\log_2$Ratio (tissue A/tissue B)]. The experiment was performed in duplicate on two different days (Exp #1, Exp #2). Correlation of LogRatios between the two experiments was >99%, demonstrating excellent reproducibility.

*Ruvkun, G. 2001. Glimpses of a tiny RNA world. *Science* **294(Oct. 26)**:797-799.

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REPORTS *CONTINUED...*

CHEMISTRY

Reversible, Metal-Free Hydrogen Activation 1124
G. C. Welch, R. R. San Juan, J. D. Masuda, D. W. Stephan
A phosphonium borate releases hydrogen upon heating above 100°C and reabsorbs it at room temperature, yielding a low-density metal-free system for H_2 storage.
>> Perspective p. 1096

GEOCHEMISTRY

Bedrock Fracture by Ice Segregation in Cold Regions 1127
J. B. Murton, R. Peterson, J.-C. Ozouf
Experiments show that rocks are fractured by the segregation and growth of ice in cracks, not by the expansion that occurs as water freezes to ice. *>> Perspective p. 1092*

ATMOSPHERIC SCIENCE

The Impact of Boreal Forest Fire on Climate Warming 1130
J. T. Randerson et al.
Boreal forest fires add to warming initially, as greenhouse gases are released, but the increased exposure of snow in burned areas produces a delayed reflection that induces cooling.

GEOCHEMISTRY

Solar Wind Neon from Genesis: Implications for the Lunar Noble Gas Record 1133
A. Grimberg et al.
Isotopes of neon from the solar wind fractionate with depth in detectors on the Genesis spacecraft; a similar process may explain enigmatic neon isotopes in lunar soils.

CELL BIOLOGY

Sara Endosomes and the Maintenance of Dpp Signaling Levels Across Mitosis 1135
C. Bökel et al.
As cells divide during development, daughter cells retain the growth signals received by their parents through equal partitioning of a subpopulation of tagged intracellular vesicles.
>> Perspective p. 1094

BIOCHEMISTRY

Abortive Initiation and Productive Initiation by RNA Polymerase Involve DNA Scrunching 1139
A. Revyakin, C. Liu, R. H. Ebright, T. R. Strick

Initial Transcription by RNA Polymerase Proceeds Through a DNA-Scrunching Mechanism 1144
A. N. Kapanidis et al.
RNA polymerase bound to DNA begins transcription by pulling downstream DNA into itself to form a scrunched intermediate that provides the force for subsequent steps.
>> Perspective p. 1097

BIOCHEMISTRY

N-Linked Glycosylation of Folded Proteins by the Bacterial Oligosaccharyltransferase 1148
M. Kowarik et al.
Bacteria can add sugars to proteins after the latter have folded, whereas eukaryotic cells do so during the protein folding process.

MEDICINE

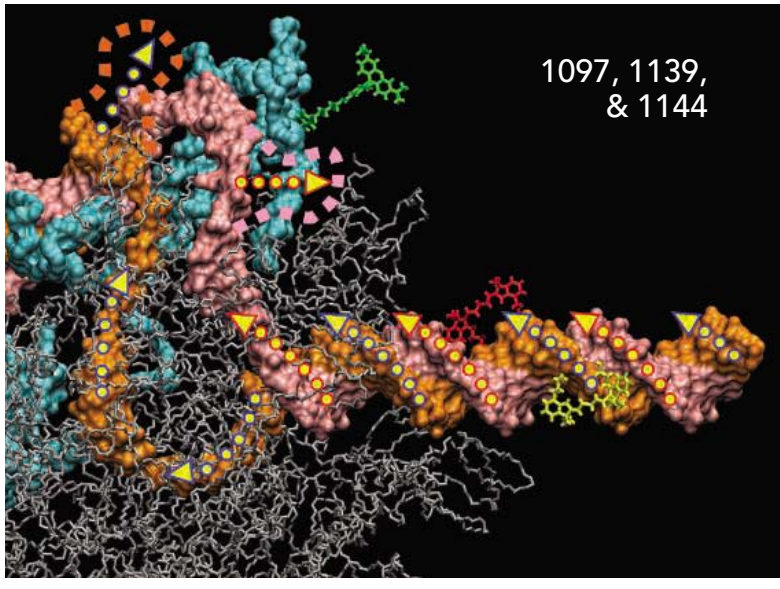
New Strategies for the Elimination of Polio from India 1150
N. C. Grassly et al.
Use of monovalent vaccines may eliminate polio from areas where poor sanitation and high population density facilitate transmission and interfere with vaccine efficacy.

PSYCHOLOGY

The Psychological Consequences of Money 1154
K. D. Vohs, N. L. Mead, M. R. Goode
In a laboratory experiment, individuals with money are less likely to seek help or offer assistance to other people.
>> Perspective p. 1091

IMMUNOLOGY

Generation of Gut-Homing IgA-Secreting B Cells by Intestinal Dendritic Cells 1157
J. R. Mora et al.
Immune cells in the gut are programmed by other cells in the nearby lymphoid tissue and a vitamin A–related signal to make antibodies that protect against gut pathogens.



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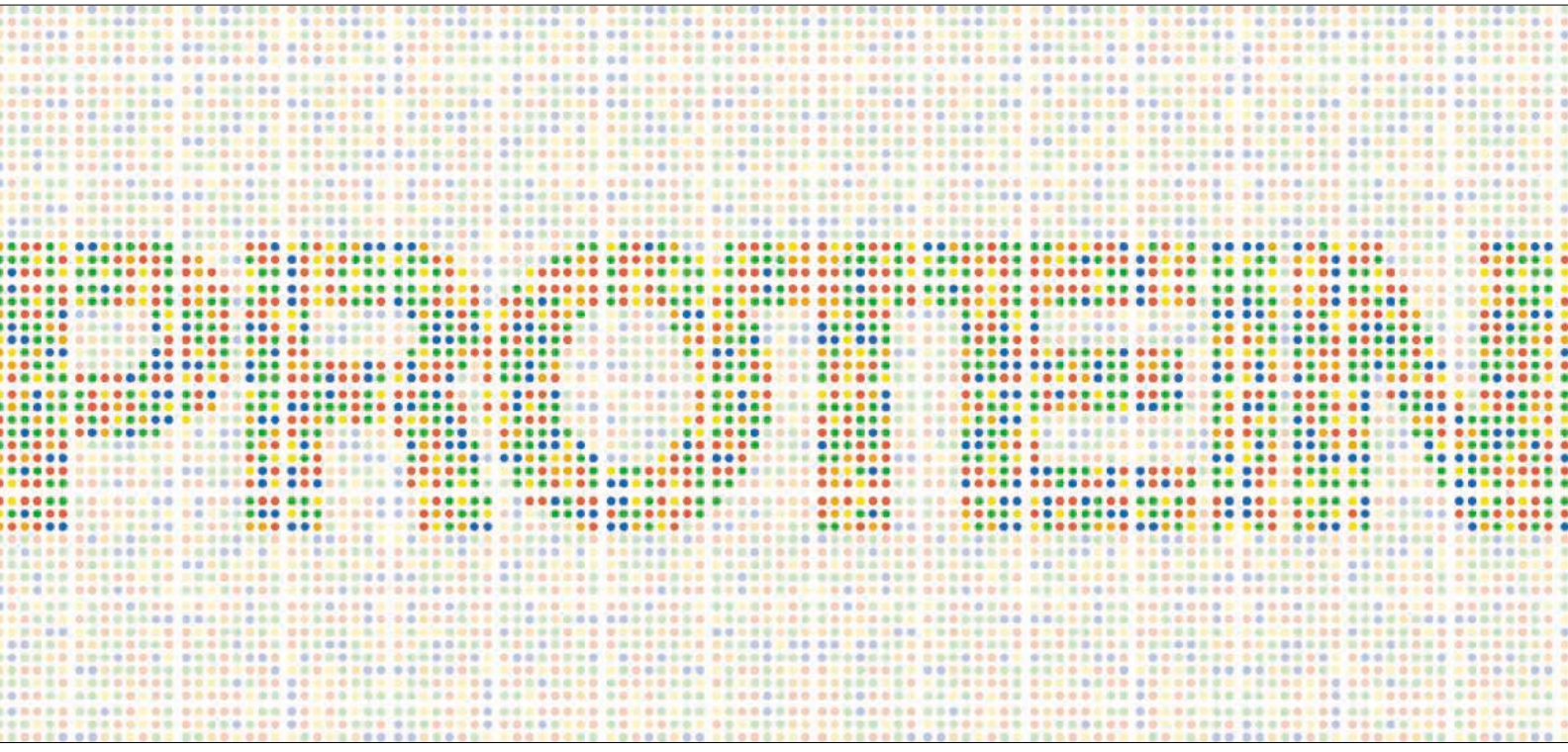
CONTENTS continued **>>**



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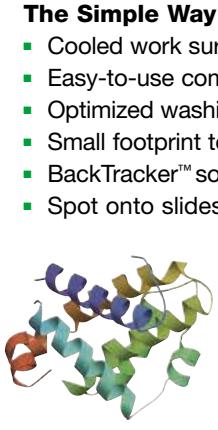

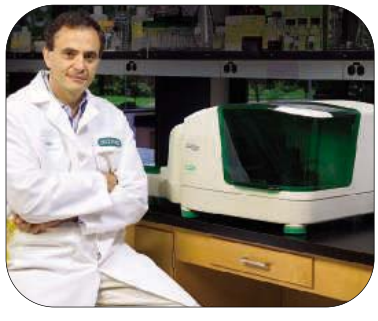

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Opportunities in high-temperature superconductivity.

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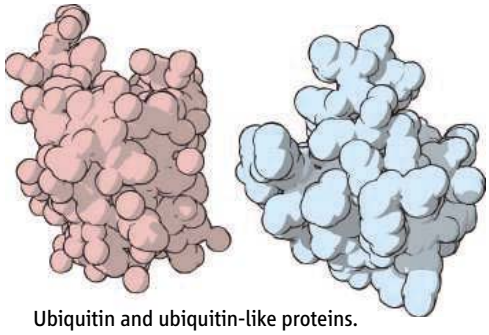
US: Tooling Up—For Love? Or Money?
D. Jensen
Some young scientists follow their hearts while others follow the money. Which is better?

GLOBAL: Special Feature—High-T_c Superconductors, Boom or Bust?
J. Austin
The expectations for high-temperature superconductors were once high but have mostly remained unfulfilled.

ITALY: A Microcosm of High-T_c Opportunities
S. Biggin
The founders of Columbus Superconductors in Genoa remain true believers in the potential of high-T_c superconductors.

US: Career Prospects in Superconductivity
A. Fazekas
Experts expect the job market in high-temperature superconductivity to heat up soon.

>> News stories pp. 1072, 1075, and 1078



Ubiquitin and ubiquitin-like proteins.

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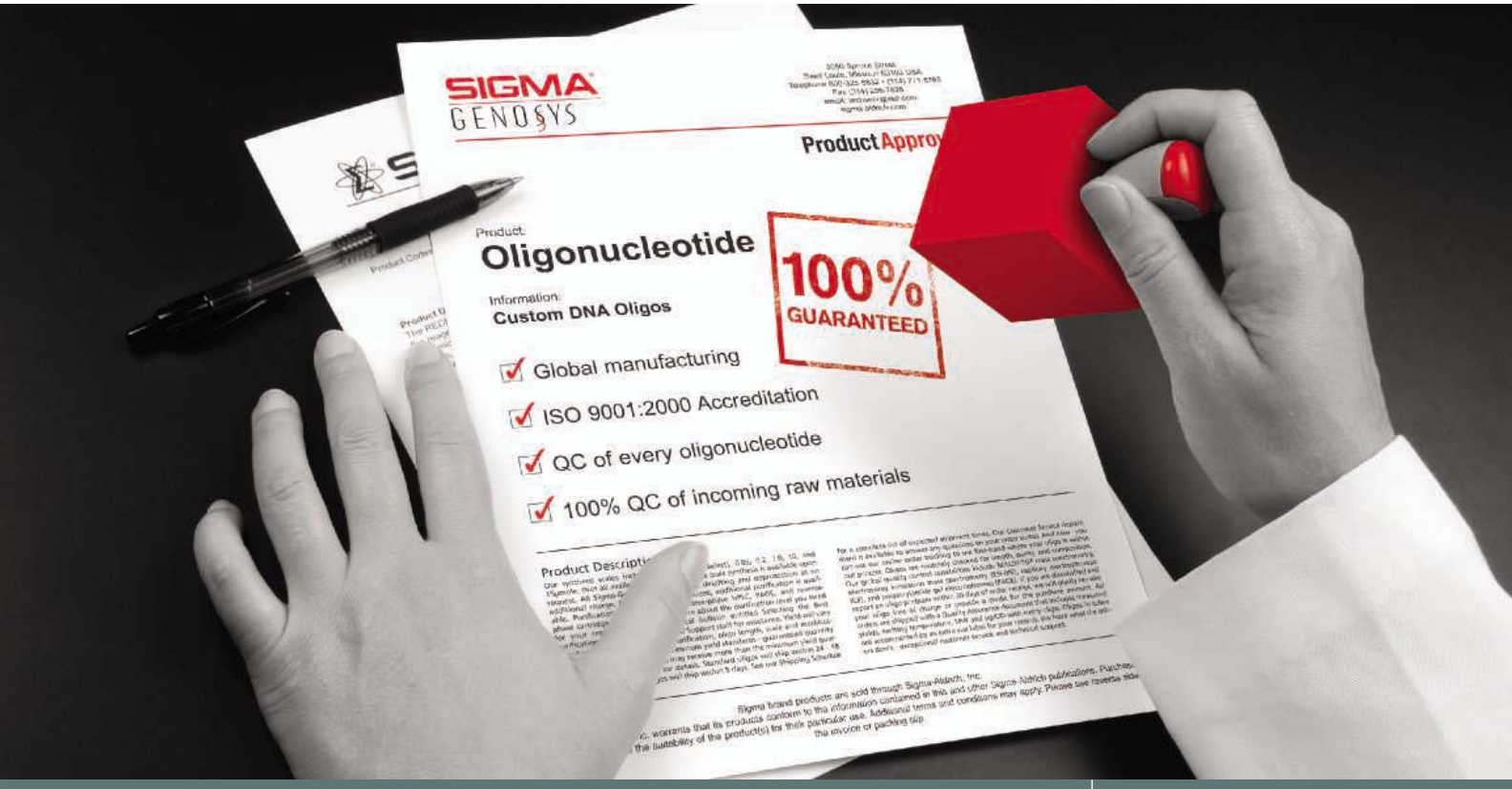
PERSPECTIVE: PTEN Regulation, a Novel Function for the p85 Subunit of Phosphoinositide 3-Kinase
D. F. Barber, M. Alvarado-Kristensson, A. González-García, R. Pulido, A. C. Carrera
The liver can adapt to loss of p85 by decreasing PTEN activity, thereby restoring insulin sensitivity.

PERSPECTIVE: Ubiquitin and NEDD8—Brothers in Arms
M. H. H. Schmidt and I. Dikic
The Cbl E3 ligase can mediate both ubiquitylation and neddylation of the epidermal growth factor receptor.

Proven!



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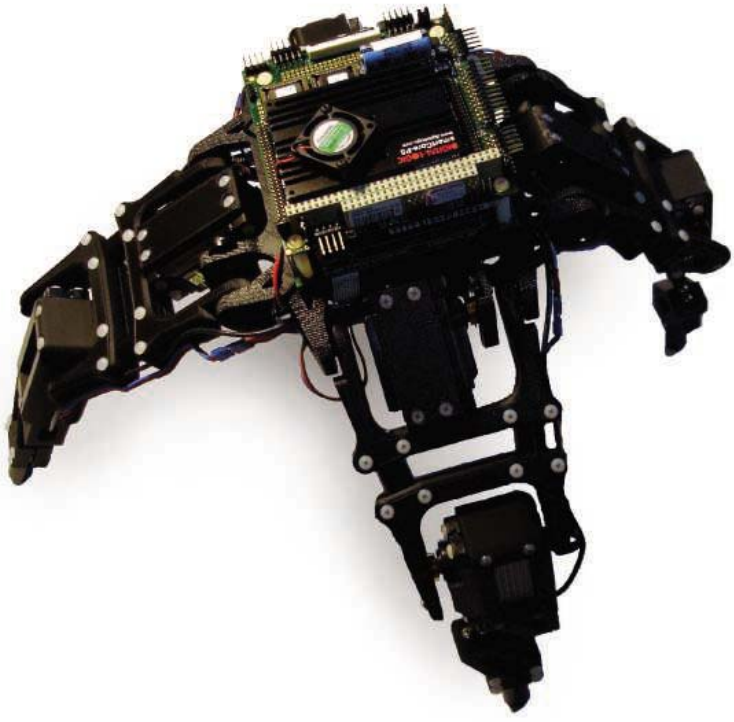


<< Damaged, Detained, But Undeterred

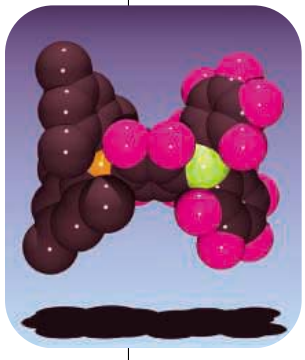
A robot may operate autonomously in controlled environments, but in new or dangerous terrain, it may become stuck or damaged. Robots are often preprogrammed to deal with specific situations, but this precaution does not help with unexpected events. **Bongard *et al.*** (p. 1118; see the Perspective by **Adami**) have constructed a robotic system that can sense and recover from damage to its structure without prior programming. The robot creates an internal model of its structure that is continually updated to account for change.

Linear Materials, Nonlinear Heat Flow

Electrical rectifiers allow current to flow in one direction, but it would seem that devices that could rectify thermal energy and direct heat flow would violate Fourier's Law. However, Peierls noted more than 50 years ago that in one dimension, heat flow can be anomalous, and recent theoretical work has suggested that rectification could be possible, albeit experimentally challenging. **Chang *et al.*** (p. 1121; see the news story by **Service**) report a rectification effect on the order of a few percent in which either carbon or boron nanotubes are given an axial asymmetry by the creation of a gradient on high-mass organoplatinum molecules at one end. The authors attribute the rectification effects to heat being carried by solitons.

P, B, and H_2

Many transition metal compounds can reversibly add and eliminate H_2, but compounds of lighter elements tend not to undergo this reaction sequence cleanly, as a result of both unfavorable bonding thermodynamics and poor orbital alignment for efficient kinetics. **Welch *et al.*** (p. 1124; see the Perspective by **Kubas**) find that an air-stable phosphonium borate compound with P–H and B–H bonds cleanly liberates H_2 above 100°C, and efficiently adds it back upon exposure in solution to the gas at room temperature. The reaction is unusual in that dimesitylphosphine adds to the para carbon of a phenyl ring in tris(pentafluorophenyl)borane, rather than binding more conventionally to the boron center. The reaction may have implications for the development of relatively light-weight substances for storage and release of hydrogen.



CREDITS (TOP TO BOTTOM): BONGARD *ET AL.*; WELCH *ET AL.*

Cold Snap

The shattering of rock by ice freeze has long been thought to be caused by volumetric expansion when water distributed within the rock freezes. However, **Murton *et al.*** (p. 1127; see the Perspective by **Hallet**) demonstrate experimentally an alternative mechanism, called ice segregation, which operates when there is a temperature gradient. As the freezing front moves through the rock, it squeezes water from its pores into pockets where ice lenses form, which causes the rock to crack. Cold-room experiments quantified this process by monitoring heave, temperature, moisture, and pore-pressure for two distinct thermal regimes. The results are verified with numerical modeling and are consistent with field observations. In warming climates, such fracturing may increasingly destabilize permafrost in polar regions.

Neon Puzzle Solved

Noble gas isotope ratios in lunar soils differ from those of the solar wind, and the explanation given has been that the lunar soils recorded a second component of energetic solar noble-gas particles that may have been stronger in the past but that is not now identifiable. **Grimberg *et al.*** (p. 1133) measured how neon in the solar wind decomposes when caught in glass detectors on the Genesis spacecraft, and they observed a change in isotope ratio with depth of implantation caused by fractionation. This process can explain the variation seen on the Moon's surface without recourse to other mechanisms.

Neanderthal Metagenomics

Our understanding of Neanderthal biology and culture remains limited. These extinct hominids are thought to have been genetically distinct from the human lineage. **Noonan *et al.*** (p. 1113; see the news stories by **Pennisi** and **Balter**) have now obtained sufficient amounts of Neanderthal genomic sequence, based on sequencing of nuclear DNA from a 38,000-year-old specimen, to create a metagenomic library. They find that humans and Neanderthals shared a common ancestor up to ~706,000 years ago and that the populations split ~370,000 years ago.

Making RNA, One Molecule at a Time

In the initial steps of transcription, RNA polymerase (RNAP) binds to promoter DNA and engages in abortive cycles of synthesis and release of short RNA products until it escapes the promoter and enters processive RNA synthesis. How RNA translocates relative to DNA in the initial transcribing complex has been controversial, with three models proposed

Continued on page 1047

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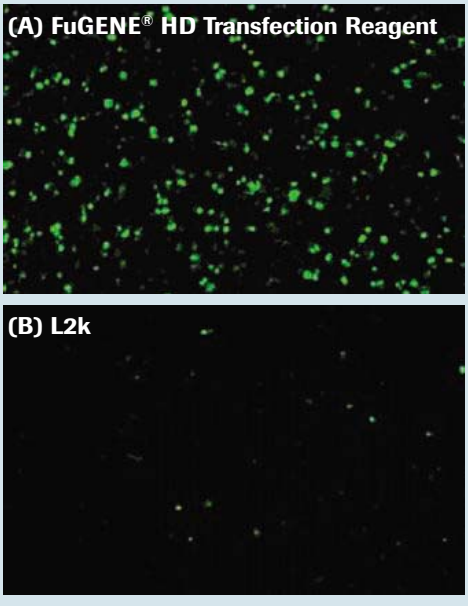

Figure 1: GFP expression in RAW 264.7 cells, 48 hours following transfection using (A) FuGENE® HD Transfection Reagent or (B) a transfection reagent (L2k) from another supplier.

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Continued from page 1045

(see the Perspective by **Roberts**). Now two single-molecule studies, one using fluorescence-energy transfer, by **Kapanidis *et al.*** (p. 1144), and the other using DNA nanomanipulation, by **Revyakin *et al.*** (p. 1139), show that initial transcription involves "scrunching," in which RNAP remains fixed on the promoter and downstream DNA into itself. Accumulated stress from DNA scrunching stress could thus provide the driving force for both abortive initiation and for promoter escape and productive initiation.

Incorporating Sugars After Protein Folding

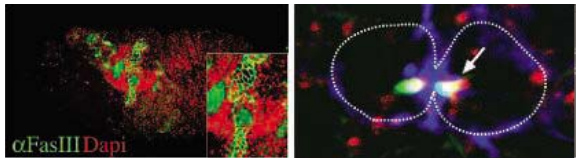
N-linked protein glycosylation is the most frequent posttranslational modification of proteins in eukaryotic cells, and a functionally homologous process also occurs in bacteria. The key component of this bacterial system is PglB, an oligosaccharylotransferase that catalyzes the transfer of the oligosaccharide to selected asparagine residues within a protein. **Kowarik *et al.*** (p. 1148) show that, unlike the eukaryotic system, the bacterial oligosaccharyl transferase can act independently of the protein translocation machinery and can glycosylate fully folded proteins in vitro.

Improving Polio Vaccine Efficacy

Critics of current plans to eradicate poliovirus have questioned whether eradication is feasible. Of the four remaining countries where polio is endemic, India represents perhaps the greatest challenge to global eradication because transmission continues despite massive immunization efforts. **Grassly *et al.*** (p. 1150) use disease surveillance data collected since 1997 to show that high population density and poor sanitation are causing persistence by facilitating the transmission of poliovirus and by severely compromising the efficacy of the live-attenuated vaccine. Switching to the monovalent form of the vaccine could potentially provide increased efficacy and allow eventual eradication.

Sorting, Signaling, and Sara

Morphogenic gradients of signaling molecules are key to tissue patterning during development. Endocytic compartments have been shown to play a role in the generation and maintenance of such gradients. **Bökel *et al.*** (p. 1135; see the Perspective by **Knoblich**) now provide evidence that the apical signaling endosome, characterized by the presence of the protein Sara (Smad anchor for receptor activation), uses the mitotic spindle to distribute developmentally important signaling molecules across division. This process ensures that the two daughter cells retain information contained in the morphogen gradient.



CREDIT: BÖKEL *ET AL.*

The Psychological Value of Money

Money can be exchanged for material goods that are essential for our physiological and psychological well-being, but are there direct effects of money on our psychological state and behavior? **Vohs *et al.*** (p. 1154; see the Perspective by **Burgoyne and Lea**) primed human subjects to think about having money and found that these subjects acted in a more self-sufficient fashion than those who were not primed. Possessing money made it less likely that subjects would ask for help in solving a problem, or offer help to another person, or make donations. In addition, subjects with money would distance themselves—literally and figuratively—from others.

Directing the Muscosal Immune Response

The mucosal lining of the intestine is stuffed with antibody-secreting B cells, which produce vast quantities of immunoglobulin A (IgA); a specialized form of antibody equipped specifically for secretion across the gut wall, where it protects against enteric pathogens. The cues that make a mucosal B cell produce IgA, rather than any of the other forms of antibody, are unclear. **Mora *et al.*** (p. 1157) now show that another immune cell, the dendritic cell, imparts this information within lymphoid tissue associated with the gut. Once activated by the gut dendritic cells, B cells become "imprinted" to enter the circulation and then home back to the mucosal lining, to begin IgA production. Induction depended on the vitamin A metabolite retinoic acid, which may explain why vitamin A deficiency exacerbates childhood diarrheal disease in the developing world.

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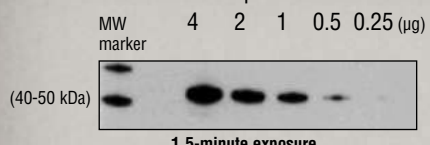

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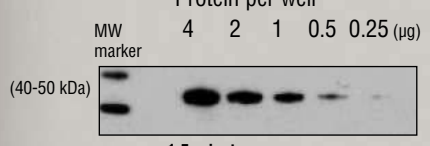

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Pierce ECL Substrate Western blot detection of Actin (Beta) from HeLa cell lysate. Dilutions of HeLa cell lysate were prepared and separated by electrophoresis. The proteins were transferred to nitrocellulose membranes (Product # 88025) and the membranes were blocked with 5% skim milk. After blocking, the membranes were incubated with Mouse Anti-Human Actin β (US Biological, Swampscott, MA) at 1 µg/ml. The membranes were washed and then incubated with 0.2 µg/ml of HRP-conjugated Goat anti-Mouse IgG (Product # 31430) and then washed again. Working solutions of the substrates were prepared according to the manufacturers' instructions and added to the membranes for 1 minute. The membranes were removed from the substrates and placed in plastic sheet protectors. Each membrane was exposed to CL-XPosure™ Film (Product # 34090) for 90 seconds.

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M. Granger Morgan is head of the Department of Engineering and Public Policy at Carnegie Mellon University, Pittsburgh, PA. E-mail: granger.morgan@andrew.cmu.edu

Don't Grandfather Coal Plants

THE UNITED STATES, THE WORLD'S LARGEST EMITTER OF CARBON DIOXIDE (CO_2), GENERATES about half of its electricity by burning coal. In terms of capacity, the average coal plant is just over 30 years old. Although most have been renovated and upgraded, there is no escaping the fact that these plants are aging, and sooner or later many will have to be replaced. Since 2002, the U.S. Department of Energy's National Energy Technology Laboratory (NETL) has tracked the plans of U.S. electricity generators to build new coal-fired power plants. They report plans to build 154 gigawatts (GW) of new coal plants over the next 24 years, and 50 GW in just the next 5 years, a big jump from the 6 GW built during the past 5 years. Age is only one reason for this expected boom in new coal plant construction. In recent years, natural gas has been the fuel of choice for new power plants. High and volatile gas prices are now contributing to what NETL calls "the resurgence of coal." However, for each kilowatt-hour generated, coal plants emit roughly twice as much CO_2 as gas plants. If the United States builds a large number of new very long-lived coal plants, reducing future emissions of CO_2 will become vastly more expensive than it needs to be.

Why should the United States limit emissions, given that other major CO_2 emitters, including China, are not doing so? Although other nations are adding coal power too, unless the United States takes action, other nations, especially industrializing nations, cannot be expected to follow.

Most U.S. utility executives believe it likely that CO_2 emission constraints will be imposed in the United States within a decade. No one knows exactly what form they will take, although economists argue for a gradually escalating tax on every ton of CO_2 emitted. But in U.S. politics, "tax" is a dirty word, so a more likely strategy is a cap-and-trade system with emission permits. Those permits will have to be allocated to start the process, and some planners of new plants may hope that their allocations will be proportional to their generators' emissions when regulation begins. Because permits will become more valuable as their numbers gradually shrink over time, that allocation scheme could hand a future windfall to firms that built substantial new capacity now. Of course, another possible approach to emission constraints would be to mandate controls only on new plants, while exempting existing plants for some extended period on the grounds that firms would otherwise face large "stranded costs." Some investors may be counting on this or on the hope that such costs could be passed on to electricity rate-payers.

Although a number of U.S. states are moving to control CO_2 emissions from power plants, federal regulation is probably unlikely for the duration of the Bush presidency. However, with the changed political complexion of the Congress, federal legislation might be possible, stipulating that when CO_2 controls are imposed, no plant built after 2006 will be exempted from coverage (that is, grandfathered), no matter what form future controls on emissions may take. Such a law would not prevent the construction of new coal plants but would strongly encourage builders of conventional coal plants to design them so as to permit amine-based CO_2 "scrubbers" to be added later. It would also provide an incentive for those building new plants to adopt advanced "clean coal" technology such as integrated gasification combined-cycle or oxyfuel plants that can capture and sequester CO_2 in deep geological formations.

Federal legislation would clearly be best. But if that is impossible, a number of state legislatures might adopt such laws. A state-by-state approach is not optimal but could clearly place future liability on investors, not rate-payers, and thus send a clear message to those planning new plants and help to create political momentum for subsequent action at the federal level.

– M. Granger Morgan

10.1126/science.1135210

Silene vulgaris.

GENETICS

The Variation Within

Uniparental (usually maternal) inheritance of a single type of mitochondrial genome, referred to as homoplasmy, has long been assumed to be the main mitochondrial state in eukaryotes. However, rare examples of multiple mitochondrial types within an individual, a state known as heteroplasmy, have been identified in animals, fungi, and plants.

Previous greenhouse studies indicated that heteroplasmy can occur in the bladder campion plant (*Silene vulgaris*), but Welch *et al.* show that it can be found at frequencies of up to 26% within a natural population. Furthermore, mothers that were heteroplasmic were shown to pass it on to their offspring, and the pattern of inheritance suggested that heteroplasmy was genome-wide (in the mitochondria) and not locus-specific. Although these findings may be taken as consistent with biparental inheritance, the fact that high levels of cytoplasmic male sterility, caused by cytonuclear interactions, are known to occur in *S. vulgaris* suggests that heteroplasmy may be selected for within female individuals in some populations. — LMZ

Genetics **174**, 829 (2006).

ECOLOGY/EVOLUTION

With Size Comes Stability

The webs of interactions between producers, consumers and decomposers in natural ecosystems confront the ecologist with a bewildering complexity. Much effort has gone into exploring the structure of food webs and the forces that contribute to their stability.

In two studies, Brose *et al.* estimate the consequences for food-web stability of the body-size distributions of consumer and resource species. Their theoretical simulations suggest that the population persistence of predator and prey species in food webs increases as the ratio of the predator-to-prey body-mass increases, up to a saturation point that is higher for vertebrates than invertebrates. These patterns were found to hold in a survey of body-size distributions in natural food webs, which also revealed that body-size ratios of predators and prey differed across freshwater, marine, and terrestrial ecosystems. These effects of body-size ratio on stability and complexity in food webs add an important dimension to the study of this fundamental ecological question. — AMS

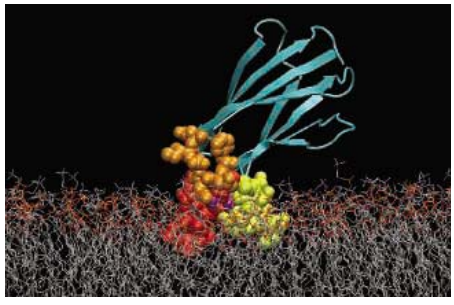
Ecol. Lett. **9**, 1228 (2006); *Ecology* **87**, 2411 (2006).

BIOPHYSICS

Dipped in Oil

The phospholipid bilayer of biological membranes is first and foremost a means of demarcating aqueous compartments by establishing a hydrophobic barrier that restricts the permeability of water-soluble compounds. One of the many additional functions of phospholipids is to provide the fatty acid substrates that can be converted into important signalling messengers, such as leukotrienes and prostaglandins. In order to cleave the linkage between the hydrophobic fatty acid and the hydrophilic headgroup, the enzyme phospholipase A2 (PLA2) attaches itself to membranes via its C2 domain, which contains binding sites for two calcium ions.

Starting from structural and biophysical constraints, Jaud *et al.* have carried out a molecular dynamics simulation of the interaction between the PLA2 C2 domain and a phosphatidyl choline bilayer. They find that the neighboring lipids reorganize to form a crater-like indentation, with the alkyl chains lining the bottom and the polar headgroups around the rim. Into this depression fit the three calcium-binding loops (CBLs) and the two complexed Ca^{2+} ions, whose primary role seems to be to mask the negatively charged loops rather than to coordinate directly to the phosphoryl oxygens, from which they are insulated by a layer of water molecules. — GJC



Docking of the CBLs (red, orange, yellow) of the C2 domain (blue).

Biophys. J. **91**, 10.1529/biophysj.106.090704 (2006).

CHEMISTRY

Zero Tolerance

In carbenes, a carbon atom engages only two of its valence electrons in bonds to other atoms, leaving the remaining two electrons free to react. Over 40 years ago, a different class of divalent carbon compound was prepared, termed a carbodiphosphorane (CDP), in which the central C was bound to two phosphines in a motif that has often been represented as cumulated double bonds: $R_3P{=}C{=}PR_3$, where R is a halide, amide, or hydrocarbon substituent. More recently, several stable free and complexed CDPs have been characterized, prompting Tonner *et al.* to explore the valence structure more thoroughly. Using quantum chemical calculations to analyze reported as well as model compounds, they find that unlike carbenes, CDPs are best described as donor-acceptor complexes: each phosphine datively donates two electrons to a central carbon, in the zero oxidation state, that has two essentially nonbonding lone pairs. The basicity of these lone pairs is borne out in a new compound, synthesized by the authors, that links two protonated CDP moieties to a silver cation. — JSY

Angew. Chem. Int. Ed. **45**, 10.1002/anie.200602552 (2006).

MATERIALS SCIENCE

Stickier with SWNTs

Adhesives that bond quickly and firmly to most surfaces on application of only a small amount of pressure are increasingly sought to eliminate the need for chemical activators or crosslinkers. Under tension, such pressure-sensitive adhesives form cavities that expand into fibrils, which in turn extend before detaching from the surface; it is these processes that contribute to the energy of adhesion. Wang *et al.* explored the adhesive properties of a poly(butyl acrylate) dispersion mixed with single-walled carbon nanotubes (SWNTs) that were functionalized with poly(vinyl alcohol) to confer hydrophilicity. They found that the SWNTs had the somewhat surprising effect of rendering the polymer both stiffer and more dissipative, two characteristics that usually vary in opposing fashion. Improved adhesive properties resulted from SWNT loading as low as 0.05 weight %, with 0.3 weight % proving optimal. During debonding, the SWNTs were found both to reduce the nucleation of cavities and to stabilize the walls between cavities, thus allowing them to absorb more energy before detachment from the substrate as fibrils. The optimized material also exhibited high optical clarity and a 10-order-of-magnitude increase in conductivity. These features bode well for eventual applications of this relatively environmentally benign material in electronics and displays. — MSL

Debonded adhesive surface.

CREDIT: WANG *ET AL.*, *ADV. MATER.* **18**, 2730 (2006)

Adv. Mater. **18**, 2730 (2006).

ASTROPHYSICS

Early Natural Selection

The bright light emitted by quasars is powered by the infall of gas toward giant black holes in galactic cores. The first quasars are known to have had central black holes that comprised a billion solar masses within a region the size of a solar system. Because assembling such a massive black hole should take billions of years, astronomers have had difficulty explaining the presence of quasars in the early (billion-year-old) universe. Volonteri and Rees have modeled the growth of the first supermassive black holes, including the competing effects of gas accretion and the dynamics of black hole mergers in their calculations. Black holes may grow by accreting gas from their surroundings, but during the most efficient accretion periods, growth is slowed by increased radiation of energy. Mergers with small companion galaxies also contribute to growth but can be destructive as well. Coalescence may be prevented if merging black holes are expelled from the galaxy by recoil from asymmetric gravitational waves and multiple-body dynamics. The authors thus frame a Darwinian natural selection scenario for black hole growth in the very young universe. Rapid and efficient growth would proceed in the highest-density regions, where gas accretion was optimal and gentle mergers outcompeted recoil losses. — JB

Astrophys. J. **650**, 669 (2006).

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<< A Complex Mode of Glucose Signaling

In *Arabidopsis*, hexokinase 1 (HXK1) acts a glucose sensor to regulate gene expression and plant growth, which is a role far removed from its function in glycolysis. However, the mechanisms whereby HXK1 mediates glucose signaling have been unclear. After showing that a small fraction of *Arabidopsis* HXK could be found in the nucleus, Cho *et al.* used proteomic and two-hybrid screens to identify two proteins—vacuolar H^+-ATPase B1 (VHA-B1) and the 19*S* regulatory particle of proteasome subunit (RPT5B)—as nucleus-specific HXK1-interacting partners that formed a complex with HXK1. Genetic analysis revealed that *vha-B1* and *rpt5b* loss-of-function mutants resembled the HXK1 *gin2* (glucose-insensitive 2 mutant): All three mutants were insensitive to repression of cotyledon expansion, chlorophyll accumulation, and leaf and root development by high glucose and showed growth retardation as compared to wild-type plants under low-light low-nutrient conditions. Like *gin2*, the *vha-B1* and *rpt5b* mutants did not exhibit glucose-mediated repression of the chlorophyll a/b–binding protein (*CAB*) and carbonic anhydrase (*CAA*) genes. Chromatin immunoprecipitation analysis showed that the HXK1 complex bound to the *CAB2* promoter; this binding was reduced but not abolished in the *vha-B1* and *rpt5b* mutants. Thus, these three proteins appear to form a complex that functions in the glucose-mediated regulation of gene transcription. — EMA

Cell **127**, 579 (2006).

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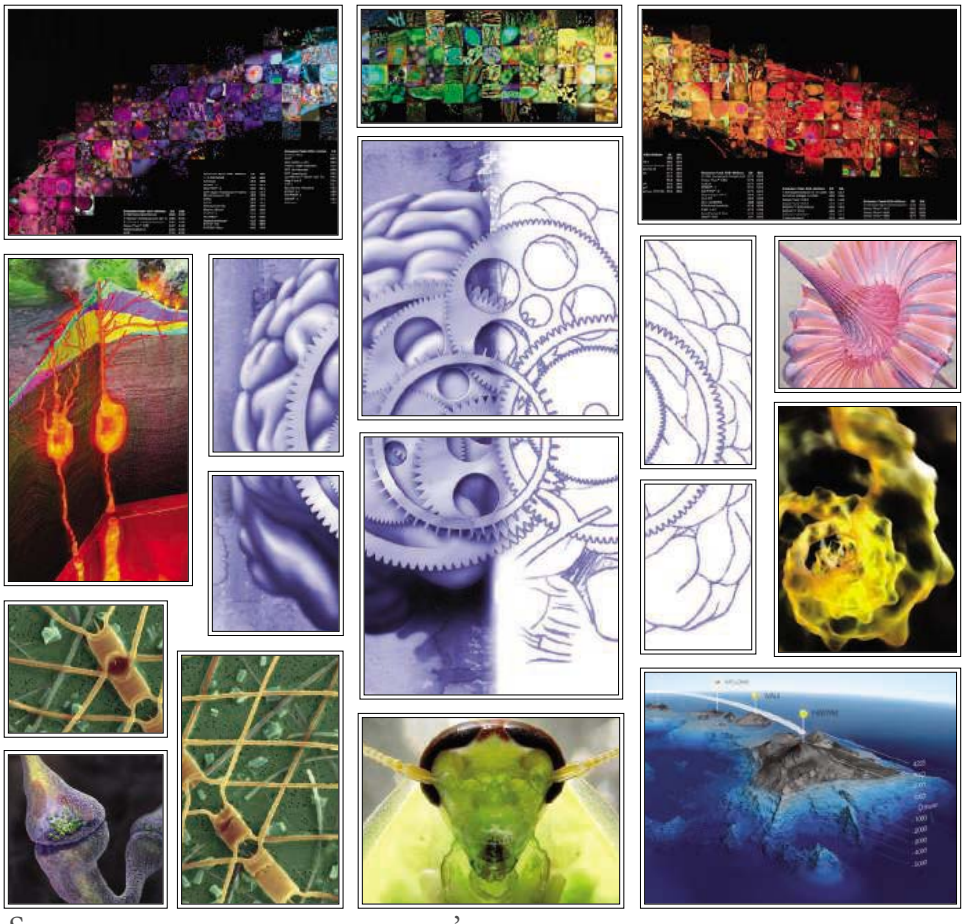

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SPACE SHOW

NASA's Jet Propulsion Laboratory in Pasadena, California, last week revealed this dazzling image that combines data from the Hubble Space Telescope and the Spitzer Space Telescope, its infrared-seeing cousin. It's an in-depth view of the Orion Nebula centered on the Trapezium, the four massive stars at its heart.

The raw data for the image were a series of numbers indicating where on the electromagnetic spectrum the light occurs. Spitzer astrophysicist Robert Hurt and his colleagues shifted the infrared wavelengths detected by Spitzer into the channels of the visible spectrum, making shorter wavelengths bluer and longer ones redder. The blues and greens in the image are from Hubble's ultraviolet and visible-light data; they show heated and ionized hydrogen and sulfur gas. The reds, oranges, and yellows are from organic molecules sensed by Spitzer. "The public is still bothered by the term 'false color,' as if there's something not quite kosher about it," says Hurt. "The colors are real; they're just beyond the perception of the human eye because they're outside the visible spectrum."

FISHY MISSING LINK?

The capture last month of a dolphin with a pair of rarely seen hind fins has electrified marine mammal researchers worldwide. "This gives us a peek at what these animals might have looked like tens of millions of years ago," says Seiji Ohsumi, a marine mammal specialist at the Tokyo-based Institute for Cetacean Research. The find, netted by Japanese dolphin hunters, may bolster theories that marine mammals returned to the sea after adapting to life on land.

Hans Thewissen, a cetacean evolution expert at Northeastern Ohio Universities College of Medicine in Rootstown, says that such limbs are rare and have previously been sighted only on dead animals: "It's a monster in some respects, but it is exciting as we've all thought the genetic programming [for such limbs] is there but switched off." Dolphin embryos have hind limbs that ordinarily disappear before birth. He says the living specimen provides unique opportunities for experiments that might help clarify evolutionary processes.

Ohsumi is collaborating with the Taiji Whale Museum in Japan on further research with the animal, which is currently in a netted enclosure in Taiji Bay, about 400 kilometers west of Tokyo.

TALES FROM THE OUTHOUSE

Evidence for an ancient latrine in Qumran, a settlement on the northwest shore of the Dead Sea in Israel, has bolstered the idea that Qumran was occupied by the Essenes, a strict, all-male Jewish sect linked to the Dead Sea Scrolls.

Some years ago James Tabor, a scholar of early Christianity at the University of North Carolina, Charlotte, spotted what appeared to be the remains of ancient toilet stalls behind a bluff about 1000 meters northwest of the Qumran camp. Recent soil samples turned up intestinal parasites specific to humans.

The find supports the notion that the Essenes did in fact inhabit Qumran from around 150 B.C.E. to 70 C.E., Tabor reports in the forthcoming issue of the journal *Revue de Qumran*. The men apparently followed toiletry practices prescribed in the scrolls, which included placement of latrines out of sight of camp and burial of feces.

Alleged latrine is behind rocks at upper left.

The latrine may also help explain why more than 90% of the men interred in a Qumran graveyard died before age 40. Burial of feces meant that intestinal parasites survived rather than being dried up in the sun, says Tabor. The men evidently tracked the pathogens into a pool they were required to immerse themselves in on returning to camp. "In effect, the pool becomes a toxic waste pool," he says.

"There is a great deal of debate among scholars about how [Qumran] functioned and who lived there," says historian Joan Branham of Providence College in Rhode Island. "The discovery of a possible latrine could be an important piece of the overall puzzle."

NETWATCH >>

Cancer Gene Cache

The first project to share cancer-promoting genes found by scanning the entire human genome has posted its initial results. The Cancer Genetic Markers of Susceptibility program, sponsored by the U.S. National Cancer Institute, evaluated DNA samples from some 1100 prostate cancer patients and an equal number of healthy men. Researchers tested more than 300,000 single nucleotide polymorphisms (SNPs) to determine which ones boost the risk for the cancer. The data, released on 19 October, include the association values for each SNP. Scientists can break down the results to discover, say, how common a particular DNA variation is among patients with fast-spreading tumors. A whole-genome analysis of breast cancer genes will follow early next year. >> caintegrator.nci.nih.gov/cgems

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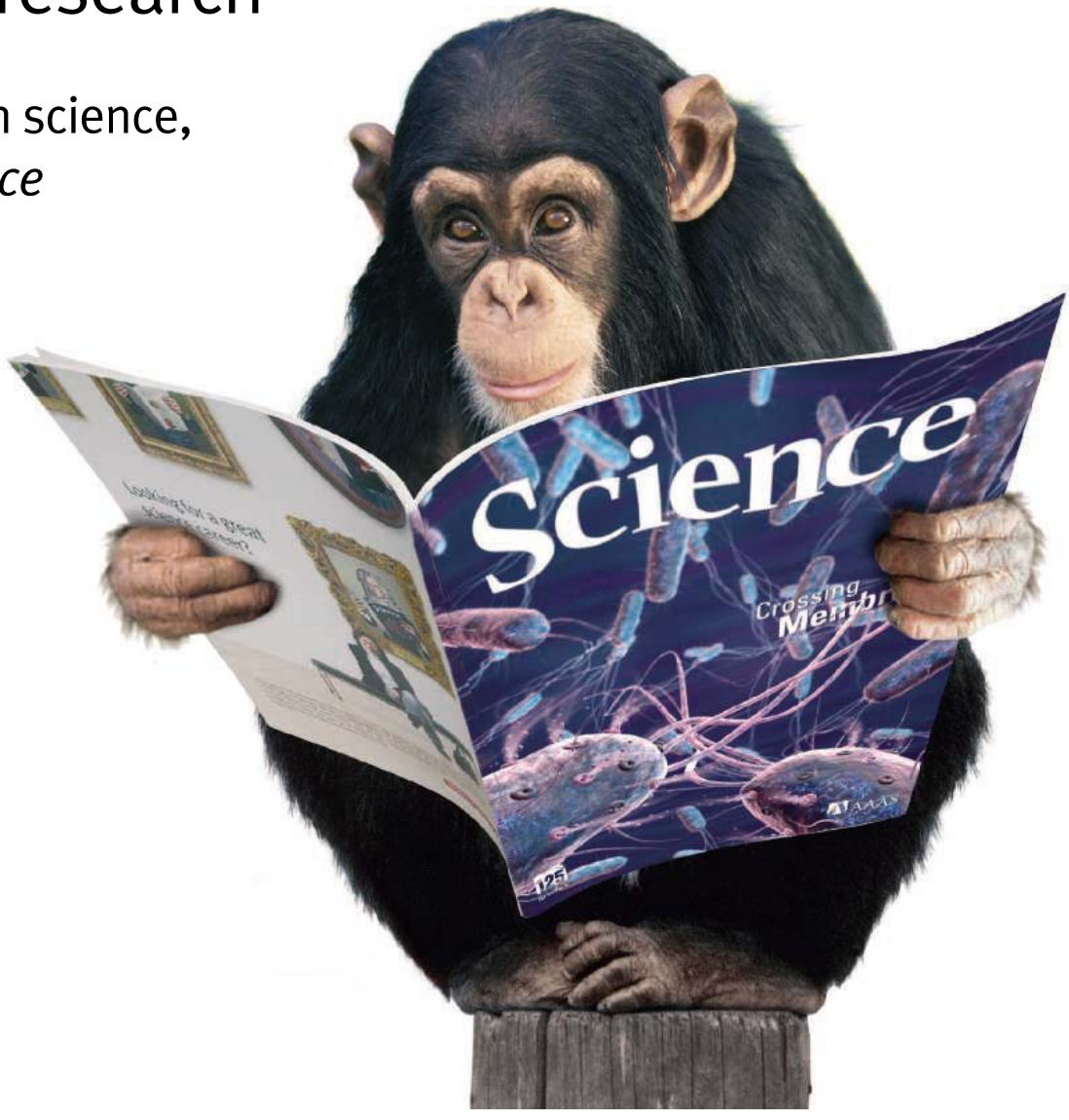

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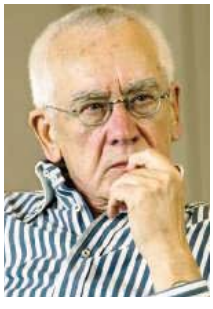
EDITED BY YUDHIJIT BHATTACHARJEE

Nonprofit World

TROUBLESHOOTER. The cash-strapped Academy of Natural Sciences in Philadelphia, Pennsylvania, has picked a new leader. William Brown, a lawyer with a Ph.D. in ecology, will take the helm in February. He replaces D. James Baker, whose 5-year contract was not renewed.

Brown, 58, once worked for environmental groups and was science adviser to the Interior Department during the Clinton Administration. Then he persuaded the world's largest trash company, Waste Management Inc., to adopt a policy of no net loss of biodiversity. He has also helped Hawaii's Bishop Museum double its endowment, to $65 million, and build a new science center.

The 194-year-old academy needs similar help. It has been running a deficit of between $500,000 and $1 million for several years. Last year, three of 10 curators were let go (*Science*, 7 January 2005, p. 28), and earlier this year, the museum sold most of its mineral collection to bolster its library's endowment. Brown says he hopes to preserve its remaining collections, renovate buildings and displays, and perhaps expand the environmental science team—largely with outside donations. Paleontologist Ted Daeschler says curators are optimistic about Brown's arrival. "He understands the scientific mission," Daeschler says. "We're very excited and hopeful."



DEATHS

IN HIS PRIME. Cancer robbed Paul Baltes of the chance to apply his theories of how best to face the challenges of old age. A director at the Max Planck Institute for Human Development in Berlin and professor at the University of Virginia, Charlottesville, Baltes died on 7 November at age 67.

Baltes showed that concentrating and honing a select skill—say, playing chess or the piano—could help compensate for the cognitive declines associated with aging. He himself had little time for relaxation: Until a few days before his death, he was planning the semi-annual meeting of an interdisciplinary group of neuroscientists, economists, demographers, and psychologists that he founded 2 years ago. He died a day before the Naples meeting started.

"He had been in charge until last weekend. It's a shock to everyone," says Jacqui Smith of the University of Michigan, Ann Arbor.

MOVERS

CHANGE AT NCCAM. The chief of the National Institutes of Health's (NIH's) controversial alternative medicine institute stepped down last week for medical reasons. Stephen Straus has led the National Center for Complementary and Alternative Medicine (NCCAM) since it was started in 1999. He has strived to steer the now–$123 million center, created by Congress to study therapies such as shark cartilage supplements, into rigorous scientific territory. "He was accomplishing it," says cardiologist David Hillis of the University of Texas Southwestern Medical Center in Dallas, a member of NCCAM's advisory council who has known Straus since medical school. But critics still question some NCCAM-sponsored clinical trials and suggest that its standards lag behind those of other NIH institutes (*Science*, 21 July, p. 301).

Straus, 59, an infectious-diseases researcher, declined comment on his health issues, but Hillis and others say he has been treated for brain cancer. He will now serve as senior adviser to NIH Director Elias Zerhouni. The center's acting director will be Ruth Kirschstein, 80, former director of NIH's general medical sciences institute and once NIH acting director.

Three Q's >>

Physicist **Serge Feneuille**, 66, was director of France's National Center for Scientific Research and CEO of Lafarge, a major building materials company. Two months ago, President Jacques Chirac appointed him chair of the new 20-member High Council for Science and Technology, which advises the government on science policy.

Q: French scientists often say the government doesn't take them seriously, and some worry that the same may happen to your council.
If I thought we wouldn't be taken seriously, I wouldn't have taken the job. It's true that French governments have neglected science and technology for about 30 years. But today, politicians acknowledge that science is an important part of our national strategy. That's something new.

Q: What's ailing French science?
We have many problems, but the biggest one is micromanagement, which makes research unattractive as a profession. We need to find a way to recruit more young people, especially young women.

Q: You know the United States well. Can French science policy makers learn anything from the U.S. system?
The American system of research funding has led to autonomy for research groups, competition, and dynamism, three things that we don't have enough of in France. That's why I think it's inevitable that France and the rest of Europe slowly evolve towards the U.S. model. I call it the standard model.

Got a tip for this page? E-mail people@aaas.org

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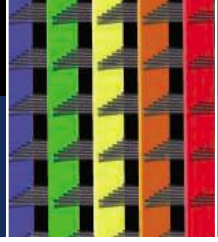


Steering phonons
1065

WHO's new boss
1067

ELECTION 2006

Science Awaits Impact of Democratic Sweep in Congress

Deal or no deal? Senate Democratic leader Harry Reid shakes hands with Vice President Dick Cheney in a meeting President George W. Bush hosted last week.

CREDIT: LARRY DOWNING/REUTERS

Science policy lobbyists like to say that strengthening the U.S. research enterprise isn't a partisan issue. That theory will be put to the test starting in January—and perhaps even sooner—when the research community tries to cash in on last week's Democratic capture of both the Senate and the House of Representatives without sacrificing expected legislative gains under the current Republican leadership.

Specific areas may benefit: Calls for relaxing constraints on embryonic stem cell research and greater environmental stewardship may have helped propel some Democrats to victory and raised hopes for action in the upcoming 110th Congress (see pages 1061, 1062). But on the overall direction of government spending on science, there's less difference between the two parties than on many issues. Both support a 2005 report from the National Academies on how to improve U.S. competitiveness—including doubling the budgets of some science agencies—for example, although they disagree on which recommendations to emphasize and how quickly to proceed. Even so, legislation to implement many of the report's suggestions has been stalled, and many lobbyists are saving their powder for the new regime.

"I don't think there's any broad message for science in the election," says Representative Vernon Ehlers (R–MI), a 13-year veteran who had hopes of chairing the House Science Committee had the Republicans remained in power. "Science continues to be largely bipartisan." Both Ehlers and Representative Rush Holt (D–NJ), who jokingly call themselves a two-person congressional physics caucus because of their Ph.D.s in the field, expect Democrats to push ahead next year with their own bills to improve U.S. competitiveness that contain major increases for research, education, and training, and clean-energy technologies. But if and when those authorization bills pass, it may be hard to find money to implement them.

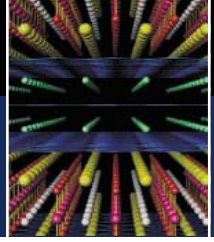
Indeed, the stage for budget battles next year could be set in the next few weeks. That's when the lame-duck Republican Congress considers appropriations bills containing hefty spending increases for several science agencies. Science lobbyists fear that some of those bills, covering the 2007 fiscal year that began 1 October and based largely on requests from President George W. Bush, could be severely trimmed to meet another goal that both parties swear allegiance to: reducing next year's expected budget deficit of $335 billion.

Although most observers are still hoping Congress will approve spending bills based on agency-by-agency negotiations, another path would be to hold every agency to 2006 funding levels under what's called a continuing resolution (CR). "A CR is the worst-case scenario," Arden Bement, director of the National Science Foundation (NSF), told a group of advisers earlier this month. "I don't want to think bad thoughts like that." An even bigger budget wild card is the direction of the war in Iraq.

The most obvious change next year will be a new lineup of committee chairs. In the Senate, that will mean a roster of familiar Democratic faces setting the scientific agenda, including Daniel Inouye of Hawaii at Commerce, Science, and Transportation; Massachusetts's Edward "Ted" Kennedy at Health, Education, and Labor; and New Mexico's Jeff Bingaman at Energy and Natural Resources. The likely new heads of research-rich Senate appropriations panels include Maryland's Barbara Mikulski (NSF, NASA, and the National Oceanic and Atmospheric Administration) and Iowa's Tom Harkin (the National Institutes of Health). All have seen their party's fortunes wax and wane and have a history of working closely with their Republican counterparts. (Only one major committee in either body will be headed by a woman: California Senator Barbara Boxer at Environment and Public Works.)

In the House, the Democratic majority will mean a return to power of well-known figures such as Michigan's John Dingell at the helm of the Energy and Commerce Committee and California's Henry Waxman at Government Reform. California's George Miller will lead the education and workforce panel, which could be busy reauthorizing programs for both elementary and secondary school students and for the nation's system of higher education. One relative newcomer will be Tennessee's Bart Gordon, in line to chair the House Science Committee (see page 1061). The heads of the science-relevant House spending panels won't be clear for several weeks. **–JEFFREY MERVIS**

Ancient DNA comes of age
1068



20 frustrating years of high-temperature superconductivity
1072

ELECTION 2006

Environmentalists See a Greener Congress

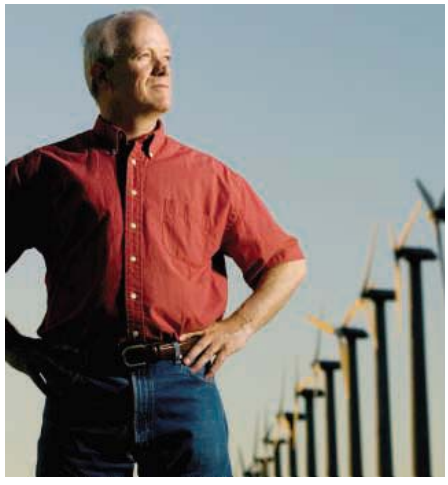
The next Congress will shift its environmental policymaking from reverse to forward, say environmental advocates celebrating last week's election results. Two major reasons for that new direction are the defeat of a powerful House member who, critics say, was bent on weakening the Endangered Species Act (ESA), and the replacement of an influential Senate chair, who infamously called global warming a hoax, with a longtime proponent of cutting emissions of greenhouse gases.

"The mood is one of excitement and anticipation," says Melissa Carey of Environmental Defense. "We haven't had a better opportunity to do something about climate change in years." The enthusiasm is tempered: Democrats are not united on the issue, have a slim majority, and face an Administration that adamantly opposes controls on emissions. Meanwhile, President George W. Bush last week asked the lame-duck Congress to pass an energy bill, fighting words for Democrats trying to block a House version that would open up much of the U.S. coastline to drilling.

The biggest news in the House was the defeat of Representative Richard Pombo (R–CA). As chair of the Resources Committee, Pombo last year won House passage of his major revision of ESA (*Science*, 7 October 2005, p. 32). The bill has since stalled in the Senate. Environmental groups contributed more than $2 million to the campaign of Jerry McNerney, a wind-power engineer, who defeated Pombo 53% to 47%, ending the attempt to rewrite the ESA.

Now environmentalists are anticipating more friendly treatment. Representative Nick Rahall (D–WV), the likely new chair, wants to reform a mining law that has led to problems with contaminated tailings, protect roadless areas in national forests, and end subsidies for offshore oil exploration. Rahall also plans to examine claims that a political appointee at the Department of the Interior distorted scientific findings to prevent the listing of endangered species.

In the Senate, California's Barbara Boxer is expected to take the helm of Environment and Public Works from Senator James Inhofe



Shifting winds. Clean-energy advocate Jerry McNerney defeated Representative Richard Pombo, who pushed for domestic oil and gas exploration.

(R–OK), a bête noire of the climate change community. Her priorities include legislation similar to her home state's that would cap and eventually reduce emissions of greenhouse gases. House Speaker–designate Nancy Pelosi is like-minded; she co-sponsored a stalled bill proposed by Representative Henry Waxman (D–CA) that would cap emissions in 2010 and then reduce them to 1990 levels over the next decade.

Such a bill would likely face resistance from Representative John Dingell (D–MI), who's slated to take over the House Energy and Commerce Committee. Dingell said last week that he would "support responsible legislation" and plans to hold hearings, but he told Greenwire that Waxman's bill is "extreme." Although some advocates complain that there's already been too much talk—239 hearings on climate change, by one count—others say that the shift in power has turned the debate from whether action is necessary to how much and when. **–ERIK STOKSTAD**

Gordon Steps Up to House Science Post

Representative Bart Gordon (D–TN) is known as the fastest man in Congress for his stellar performances each year in a 5K race that pits federal officials against the members of the media who cover them. Starting in January, however, the 57-year-old lawyer expects to be leading a slower-moving pack: the House Science Committee.

Although the science committee is little known outside the research and academic communities, Gordon says that he asked to be on it as a freshman and that "it was my hope all along" to become its chair some day. As the highest-ranking Democrat on the committee since 2003, he's all but guaranteed the job in the 110th Congress.

Winning races. Incoming science chair Representative Bart Gordon, center, also excels as a runner.

First elected in 1984 after holding Democratic Party posts in Tennessee, Gordon has been returned 11 times, mostly by comfortable margins. He succeeded Al Gore, whose election to the Senate that year launched a national career that would ▶

CREDITS (TOP TO BOTTOM): MCNERNEY FOR CONGRESS COMMITTEE; ROBERT BURGESS

take him within a hanging chad of the White House. Gordon, who still lives in his hometown of Murfreesboro, holds no such grand political ambitions, say those who have followed his career. But he still wants to make a difference. "He's a totally local politician," says Jeff Vincent, the Washington, D.C.–based head of federal relations for Vanderbilt University in neighboring Nashville. "I think this is really an opportunity for him to play a larger role."

As chair of the committee's space panel in the early 1990s, Gordon developed an interest in space-related issues that is likely to translate into closer scrutiny of the Bush Administration's proposed moon-Mars exploration program and its impact on space science. "I think that both are underfunded," he says, "but I think we need to know more before we can move ahead."

His supportive but questioning attitude toward NASA mirrors the view of the outgoing chair, retiring moderate New York Republican Sherwood "Sherry" Boehlert. In fact, the two men see eye to eye on most issues before the committee—notably, additional funding for science education at the National Science Foundation (NSF), criticism of the Administration's attempts to muzzle federal scientists on sensitive topics such as climate change, and doubling federal spending for research in the physical sciences. "I can't think of a better relationship between a chair and a ranking [minority] member than between Bart and myself," says Boehlert.

Even so, that bipartisanship may be put to the test in the next Congress. Gordon is eager to set up an entity within the Department of Energy (DOE) modeled after the Defense Advanced Research Projects Agency. Although the idea comes from an acclaimed 2005 National Academies report on strengthening U.S. science that the Administration has embraced, President George W. Bush pointedly omitted any new DOE agency from the competitiveness plan he submitted to Congress earlier this year. Gordon's desire to give NSF a bigger role in science education may also irritate the White House, which wants the Education Department in the driver's seat. And Gordon's promise to hold hearings "to give scientists a chance to tell their side of the story" about whether the Bush Administration has undermined scientific integrity is sure to draw fire from Republican colleagues. **–JEFFREY MERVIS**

Stem Cell Supporters Hail Results, But Political Lessons Aren't Clear

A helping hand. Expanding stem cell research was a winning issue for Wisconsin Governor Jim Doyle (with actor Michael J. Fox).

CREDIT: MORRY GASH/AP PHOTO

On 7 November, voters in several states backed candidates supporting expanded research with embryonic stem cells. That much is clear. But the impact of those victories on federal policy that restricts the use of stem cells is much harder to discern. And experience in at least one state suggests that injecting stem cell issues into a political campaign can backfire.

"Republican candidates aren't going to want this as an issue in 2008," asserts Sean Tipton of the Coalition for the Advancement of Medical Research in Washington, D.C. He says the election results bolster the hopes of those seeking to overcome President George W. Bush's opposition to allowing research on embryonic stem cell lines created after August 2001. (Bush had vetoed such a bill, H.R. 810, last summer, and supporters were unable to override it.) But opponents of embryonic stem cell research take heart from the fact that they almost defeated a proposed constitutional amendment in Missouri that would bar lawmakers from outlawing the research while banning reproductive cloning; as recently as September, the proposal enjoyed a 20-point lead.

The Missouri vote has reinforced one tenet of faith among supporters: Don't make stem cell research a partisan issue. Despite her personal support for Amendment 2, Senate Democratic challenger Claire McCaskill had avoided the topic during campaign appearances out of concern about offending rural, pro-life supporters. But in the waning days of her race against incumbent Republican Jim Talent, McCaskill aired a television advertisement featuring movie star Michael J. Fox, visibly afflicted by Parkinson's disease. Fox accused Talent of voting to "criminalize the science." The ad did not mention the amendment, but it turned out to be a disaster for the amendment's supporters. National conservative icon Rush Limbaugh complained that McCaskill was trying to "mislead voters," and Fox News host Bill O'Reilly attacked philanthropists and cancer survivors Jim and Virginia Stowers of Kansas City for standing "to make billions" off various research institutions they have set up if the amendment passed—a charge that Stowers Institute President William Neaves called "outrageous."

"This became the center of the culture war universe," says Bob Deis, a political consultant to amendment backers. Internal polling showed Republican support for the amendment plummeting "eight to 10 points" in a week, says Deis. In the end, Amendment 2 passed by only 50,000 votes among 2 million cast. (It's not clear whether the ad had any effect on the Senate race itself, which concentrated on the Iraq war and health care. McCaskill won narrowly after trailing Talent for much of the campaign.)

In states awash in a stronger Democratic tide, some candidates did effectively leverage local scientific and commercial interest in the research. Incumbent Wisconsin Governor Jim Doyle, a Democrat, vetoed a bill in 2005 that ▶

would have criminalized somatic cell nuclear transfer—popularly known as research cloning—a potential method of obtaining embryonic stem cells genetically matched to a patient. After a poll showed that 69% of Wisconsin voters approved of the research, Doyle ran harder than any other U.S. candidate on the issue against an opponent—Republican Representative Mark Green—who opposed the method. In a series of press conferences and TV ads, flanked by patients and entrepreneurs, Doyle touted the proposed $375 million Institutes for Discovery and efforts to recruit stem cell experts to the state. Doyle defeated Green by 53% to 45%.

In Maryland, Democratic Representative Ben Cardin also effectively trumpeted his support for embryonic stem cells in TV ads featuring Fox. The ads claimed that Cardin's opponent, Republican Lieutenant Governor Michael Steele, shared Bush's opposition to the research. When Steele's sister proclaimed in an ad that her brother "does support stem cell research," three stem cell scientists at Johns Hopkins University in Baltimore, Maryland, held a press conference to clarify that Steele only supported work with adult cell lines. Cardin won by 54% to 44%.

Pro–stem cell lobbyists say Bush's growing unpopularity gives some Republicans a chance to vote their consciences, with less fear of political repercussions, if a stem cell bill comes up in Congress in the next 2 years. Representative Heather Wilson (R–NM), who was narrowly reelected last week in a campaign that focused on the Iraq war, explained in a TV ad that she voted to override the veto because it "was the right thing to do." Tipton says the new Democratic majority in both houses also gives proponents a chance to apply new tactics, including connecting stem cells to hard-to-veto bills, or pairing it with other legislation that appeals to pro-life lawmakers or the White House.

But David Prentice of the Family Research Council in Washington, D.C., which opposes any change in current federal policy, sees no "sea change" on the issue and predicts that supporters won't find it easy to overcome another Bush veto. And although most of the country's attention is shifting to Washington, Missouri may still bear watching. There's already talk among Missouri's pro-life community about crafting a new ballot initiative that would repeal Amendment 2. **–ELI KINTISCH**

Scientists Get Out the Word

U.S. scientists hardly play any organized role in influencing elections. But two new groups are claiming some credit for the outcome of a few races last week and say they plan to be more active in 2008.

Scientists and Engineers for America (SEA), founded in September by Nobelist Peter Agre of Duke University in Durham, North Carolina, and others, visited a handful of college campuses to support candidates favoring embryonic stem cell research, the teaching of evolution, and policies to stem global warming. The 6500-member group, which raised $95,000, also ran a few Internet banner ads and posted information on its site (www.sefora.org) to help voters see the track records of different congressional candidates on key scientific issues. Senate Democratic candidates favored by SEA won in Missouri, Maryland, and Virginia.

In Ohio, a group calling itself Help Ohio Public Education (HOPE) persuaded former U.S. representative and Akron mayor Thomas Sawyer to run in a state school board race against Deborah Owens Fink, a supporter of intelligent design. "The idea behind HOPE was in part to do what the creationists have been doing: recruiting candidates and then helping them get elected," says physicist Lawrence Krauss of Case Western Reserve University (CWRU) in Cleveland, who organized the group. Krauss also collected signatures from nearly 90% of CWRU's science faculty in support of Sawyer and four other pro-science school board candidates. "If the enemies of science can do that, why can't scientists?" he says.

Although HOPE did not raise and spend any money, it invited Brown University biologist Kenneth Miller to give public lectures about why Ohio voters needed to keep religion out of the science classroom. Sawyer trounced Owens Fink by a two-to-one margin, and three of the other four candidates endorsed by HOPE won.

Both groups plan to continue their work. SEA hopes to establish student chapters at universities and allow members to post information about where politicians stand on science. "What this election told us is that issues of science do connect with the public," says Susan Wood, former director of the Office of Women's Health at the U.S. Food and Drug Administration and an SEA founder. "Voters are becoming increasingly aware that competent governance requires making policies based on good science." **–YUDHIJIT BHATTACHARJEE**

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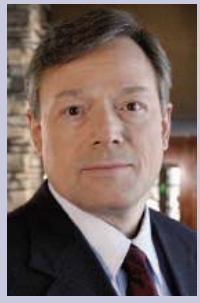
Elsewhere on the Election Front



Scientifically inclined. Wisconsin Democrat **Steven Kagen**, who won an open House seat, is an assistant clinical professor of allergy and immunology at the Medical College of Wisconsin in Milwaukee. The physician owns four allergy clinics and also maintains a lab that has published molecular analyses of several environmental allergens.

Kansas Democrat **Nancy Boyda**, who defeated five-term Representative Jim Ryun, worked as a field inspector and analytical chemist for the Environmental Protection Agency and held management positions at pharmaceutical companies. She holds an under graduate degree in chemistry and education and has taught middle-school chemistry.

Political powerhouse. Tiny Cornell College in Mount Vernon, Iowa, can lay claim to two incoming Democratic House members: political science professor **David Loebsack**, who toppled 15-term incumbent Jim Leach, and **Chris Carney**, who graduated in 1981 with degrees in environmental science and diplomatic history and now teaches political science at Pennsylvania State University, Worthington-Scranton.

Raising his voice. New York Democrat **John Hall**, who beat Representative Susan Kelly, studied physics at the University of Notre Dame in Indiana and Loyola College in Baltimore, Maryland, before dropping out to become a rock musician. A member of the popular band Orleans in the 1970s, Hall led efforts to fight nuclear power plants before turning to politics.

2008 is really open. For the first time since 1928, neither the incumbent president nor vice president will be running for president in 2008.

GLOBAL CLIMATE CHANGE

False Alarm: Atlantic Conveyor Belt Hasn't Slowed Down After All

Fitful flow. Instruments arrayed across the North Atlantic have found surprisingly variable currents that mask any slowing of the Atlantic conveyor.

A closer look at the Atlantic Ocean's currents has confirmed what many oceanographers suspected all along: There's no sign that the ocean's heat-laden "conveyor" is slowing. The lag reported late last year was a mere flicker in a system prone to natural slowdowns and speedups. Furthermore, researchers are finding that even if global warming were slowing the conveyor and reducing the supply of warmth to high latitudes, it would be decades before the change would be noticeable above the noise.

The full realization of the Atlantic's capriciousness comes with the first continuous monitoring of the ocean's north-south flows. In March 2004, researchers of the Rapid Climate Change (RAPID) program moored 19 buoyant, instrument-laden cables along 26.5°N from West Africa to the Bahamas. A few months later, they steamed along the same latitude, lowering instruments periodically to take an instantaneous "snapshot" of north-south flows. While waiting for the moored array to produce long-term observations, physical oceanographer Harry Bryden and his team at the National Oceanography Centre in Southampton, U.K., compared the 2004 snapshot with four earlier instantaneous surveys dating back to 1957. They found a 30% decline in the northward flow of the conveyor (*Science*, 2 December 2005, p. 1403), sparking headlines warning of Europe's coming ice age.

The first year of RAPID array observations has now been analyzed, and the next European ice age looks to be a ways off. At a RAPID conference late last month in Birmingham, U.K., Bryden reported on the first continuous gauging of conveyor flow. Variations up and down within 1 year are as large as the changes seen from one snapshot to the next during the past few decades, he found. "He observed a lot of variability," says oceanographer Martin Visbeck of the Leibniz Institute of Marine Science at the University of Kiel in Germany, who attended the meeting; so much variability that "more than 95% of the scientists at the workshop concluded that we have not seen any significant change of the Atlantic circulation to date," wrote Visbeck in a letter to the British newspaper the *Guardian*.

Although the immediate threat has evaporated, a difficult challenge has taken its place. "Scientific honesty would require records for decades" in order to pick out a greenhouse-induced slowing, says physical oceanographer Carl Wunsch of the Massachusetts Institute of Technology in Cambridge. "How do you go about doing science when you need decades of record?" For their part, RAPID researchers will be asking for funding to extend array operations to a decade, says Bryden. Then some combination of government agencies would have to take on the burden of decades of watchful waiting.

–RICHARD A. KERR

CHINESE DRUG RESEARCH

Novartis Invests $100 Million in Shanghai

SHANGHAI—Big pharma companies and China may not love each other for the same reasons, but relationships are blossoming. Companies are enamored of the low operating costs and the large market potential in China, whereas local officials are aflutter over foreign investment and know-how. So far, however, few big companies have moved their R&D efforts to Chinese soil (*Science*, 29 July 2005, p. 735). Many are content with long-distance relationships, outsourcing specific steps in the drug discovery process. But Novartis, the Swiss drug giant, is making a serious commitment.

Last week, Novartis unveiled plans to build a $100 million R&D center in Shanghai, a fast-growing hub of biomedical excellence. The company intends to hire some 400 mainly local scientists to focus initially on infectious causes of cancer such as hepatitis B virus, linked to a high rate of liver cancer in China. The first of two facilities is slated to open next spring. The R&D center "will encompass all stages of drug development, from early discovery all the way to clinical trials," says Novartis spokesperson Jeffrey Lockwood.

Pharmaceutical scientists in Shanghai welcome the venture. "It's a really good thing," says Zhuohan Hu, president of the Research Institute for Liver Diseases, a company that is negotiating an alliance with Pfizer. Hu and others predict that it will not be easy for Novartis to assemble and train such a large scientific workforce.

New chief. Novartis research manager En Li has been tapped to direct a planned staff of 400 scientists.

But for Novartis, China is not virgin territory. It set up an office in Beijing in 1997 and has R&D alliances with WuXi PharmaTech and the Shanghai Institute of Materia Medica, among others. Novartis manufactures one product in China that it developed with Chinese partners: Coartem, an antimalaria drug derived from wormwood based on traditional Chinese medicine.

In the past, companies have often formed task-specific partnerships to reduce the risk of renegade employees running off with a hot discovery. ▶

CREDITS (TOP TO BOTTOM): THE NATURAL ENVIRONMENT RESEARCH COUNCIL

Companies such as Merck and AstraZeneca, for example, have contracted out specific jobs to different Chinese organizations. In 2002, Novo Nordisk was the first to establish a research facility in China. It set up a small R&D shop near Beijing; Roche followed in Shanghai in 2004. Novartis, however, would have by far the biggest research investment. Lockwood downplays the risk of Novartis findings being spirited out the back door. "We see the trend improving toward more rigorous intellectual-property protection," he says.

Novartis has tapped En Li to be research director of the center, which will be down the road from Roche in Shanghai's Zhangjiang High-Tech Park. Li, a Shanghai native, joined Novartis in 2003. He's currently a research chief at the Novartis Institutes for BioMedical Research in Cambridge, Massachusetts.

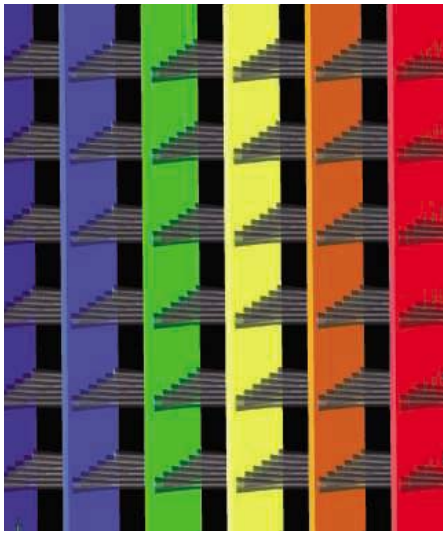
One of Li's initial challenges is to find the right mix of scientists. Although China is teeming with skilled chemists, Hu contends, "it's not that easy to find good hands-on biologists here." Lockwood is bullish. "We believe there is a growing talent pool in China," he says. "We also hope that the center will be a magnet for [returning Chinese scientists] as well." And Novartis won't be hiring all 400 scientists in one go: Its first Shanghai lab, expected to open in May 2007, will employ about 160 researchers. Construction on a second facility is planned to begin next summer.

–RICHARD STONE AND HAO XIN

PHYSICS

Electronic Nuisance Changes Its Ways

In modern electronics, as in James Bond movies, it's the good guys versus the bad guys. The good guys are electrons, packets of electrical charge that devices such as diodes and transistors start, stop, and steer to orchestrate a dance of 1's and 0's. The bad guys: vibrations called phonons that splay heat every which way and can ultimately wreak havoc on a computer chip. But now researchers at the University of California, Berkeley, may turn some unruly phonons into allies.



Hot stuff. Hypothetical material based on tapered nanotubes pushes heat from left to right.

CREDIT: ZETTL RESEARCH GROUP, UNIVERSITY OF CALIFORNIA, BERKELEY, AND LAWRENCE BERKELEY NATIONAL LABORATORY

On page 1121, researchers led by physicist Alex Zettl and mechanical engineer Arunava Majumdar report the first-ever set of simple devices, akin to diodes, that steer a small excess of phonons in one direction. "It's a cool result," says James Heath, a chemist and nanoelectronics expert at the California Institute of Technology in Pasadena. If the effect can be improved, it could lead to a novel form of computation based on phonons and to heat-steering materials that make buildings more energy-efficient, among other things.

The new work marks the latest example of the unique capability of nanostructures to display odd quantum-mechanical properties. Nanotechnologists have shown that materials with at least one dimension smaller than 100 billionths of a meter can have odd optical, electrical, and catalytic behaviors due to the way they confine electrical charges. More than 50 years ago, German-born British theoretical physicist Rudolf Peierls suggested that string-shaped one-dimensional (1D) systems could also channel heat-generating phonons in unusual ways. But researchers had never managed to demonstrate any such effect.

The Berkeley team started with tiny straw-like nanotubes, some made from carbon, others of an alloy of boron and nitrogen. In previous studies, Zettl's group and others had shown that both types of nanotubes are excellent heat conductors and that phonons move through them with equal efficiency in both directions. But Zettl's graduate student Chih-Wei Chang had been studying how phonons move through nanotubes and suspected there was an easy way to give them a push. Theoretical models suggested that a 1D system loaded with extra mass at one end would make it easier for phonons to travel from the high-mass end to the low-mass end. ▶

Agape Over Rules

Pending new rules on animal experimentation have led researchers from 33 organizations to create the European Coalition for Biomedical Research (ECBR). The coalition, announced last week in Brussels, is anticipating that the European Commission may seek to put further restrictions on the use of animals in research, through proposals such as requiring non-human primates to have been bred in captivity for at least two generations. Such a rule would have "a dramatic effect" on research, says ECBR Secretary General Mark Matfield.

Meanwhile, this week the U.S. Congress completed action on legislation that would protect the suppliers for animal research from animal-rights "terrorism." The bill, passed by the Senate in September and by the House of Representatives this week, is expected to be signed shortly by President George W. Bush.

–MARTIN ENSERINK

Chinese Flu Goes West

China's Ministry of Agriculture has agreed to let international scientists analyze 20 H5N1 avian influenza samples collected from poultry in 2004 and 2005. The samples were sent last week to a U.S. lab affiliated with the World Health Organization (WHO). A recent paper in the *Proceedings of the National Academy of Sciences* speculated about a new strain (*Science*, 10 November, p. 905) and led WHO to criticize China for not cooperating with international health organizations. The issue cuts both ways, however: WHO officials have acknowledged two cases of Western scientists failing to credit Chinese scientists for their contributions. WHO is now hopeful of getting samples on a regular basis.

–DENNIS NORMILE WITH HAO XIN

A Crewed Idea

The Indian Space Research Organisation (ISRO) is seeking government approval for its plan to send an astronaut into space. Last week, ISRO presented its plans for a manned $3.75 billion low-Earth-orbit mission by 2014 to a meeting of Indian researchers in Bangalore. The first step would be the January 2007 launch and recovery of a 525-kilogram unmanned capsule, followed by a 2008 robotic moon mission.

ISRO Chair G. Madhavan Nair says his country is not in a space race with China, which is planning a robotic lunar mission for 2007. But Nair says the move will prevent India from being "left behind" internationally. Astrophysicist Yash Pal, however, warns that "manned space missions don't do good science."

–PALLAVA BAGLA

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To test the idea, the Berkeley researchers placed individual tubes inside a vacuum chamber and bonded the two ends to a pair of custom-designed electrodes that could serve as both heaters and heat sensors. Next, they sprayed a vaporized platinum compound, $C_9H_{16}Pt$, into the chamber and used a beam of electrons from a scanning electron microscope to weld molecules of the gas onto one end of their nanotubes. They then sent a power surge with a known amount of energy to the heater and tracked how much heat made it through the nanotube to the sensor. Finally, they repeated the experiment with the heater and sensor reversed.

In every case, more heat flowed toward the side of the nanotube with less mass, even though the excess $C_9H_{16}Pt$ didn't span the two electrodes and thus couldn't carry the extra heat. Zettl suspects that standing waves called solitons that vibrate through the nanotubes could be responsible for increasing the heat-carrying efficiency in one direction, although more work needs to be done to confirm this.

For now, the effect is small. At most, only a 7% excess of phonons travels in the preferred direction. That may not be enough to create phonon-based computing devices or other applications, Heath says. But such applications "may exist if someone can figure out how to do this well," he adds. **–ROBERT F. SERVICE**

PUBLIC HEALTH

SARS and Bird Flu Veteran to Take WHO Helm

Margaret Chan is no stranger to public health emergencies. The infectious-disease expert, who was elected on 9 November to be the next director-general of the World Health Organization (WHO), is best known for her role in containing two fast-spreading outbreaks—of bird flu and SARS—as Hong Kong's director of public health from 1994 to 2003. Largely on those merits, she was awarded the top slot for communicable diseases at WHO in 2005.

But Chan says that two broader problems will be her top concerns when she takes over leadership of WHO in January. "I want to be judged by the impact we have on the health of the people of Africa and the health of women," she told the World Health Assembly just hours after being elected.

A watchful eye. Margaret Chan has experience in pandemic preparedness.

CREDIT: PETER WILLIAMS/WHO

The sudden death in May of then–Director-General Lee Jong-wook led to a hard-fought race among an unprecedented 13 nominees (*Science*, 15 September, p. 1554). Most, including Chan, had slick Web sites and spent the last 3 months campaigning around the world. From the start, Chan was among the predicted favorites, and in the final ballot she received 24 votes; the runner-up, Mexican Health Minister Julio Frenk, received 10. She will be the first Chinese to head a major United Nations organization, and many observers hope her election will encourage China's government to take a more active role in tackling international health issues such as HIV/AIDS and bird flu.

Scientists who have worked with Chan to try to prevent a global flu pandemic immediately praised her selection. "She is a very strong leader, and translating science into policy is one of her strong points," says Albert Osterhaus, a virologist at Erasmus University Medical Center in Rotterdam, the Netherlands. "In crisis situations, she knows how to handle things and how to maneuver through a political minefield."

In 1997, when the first human cases of the H5N1 avian influenza strain were detected in Hong Kong, Chan quickly responded by ordering the culling of all 1.5 million poultry on the island, an aggressive move widely credited with preventing a broader outbreak. She received more mixed reviews for her handling of the 2003 SARS outbreak; some critics say she could have pushed harder to get information from mainland China, where the disease apparently originated.

In the past few weeks, global health officials have again accused China of withholding data—this time, on the spread of avian influenza (*Science*, 10 November, p. 905). Hours after her election, Chan moved to dispel fears that she might not be tough enough on her own government. As director-general, her loyalty belongs to all 193 member countries, she said at a press conference. If anything, she said, she will be uniquely placed to encourage more openness from Chinese officials.

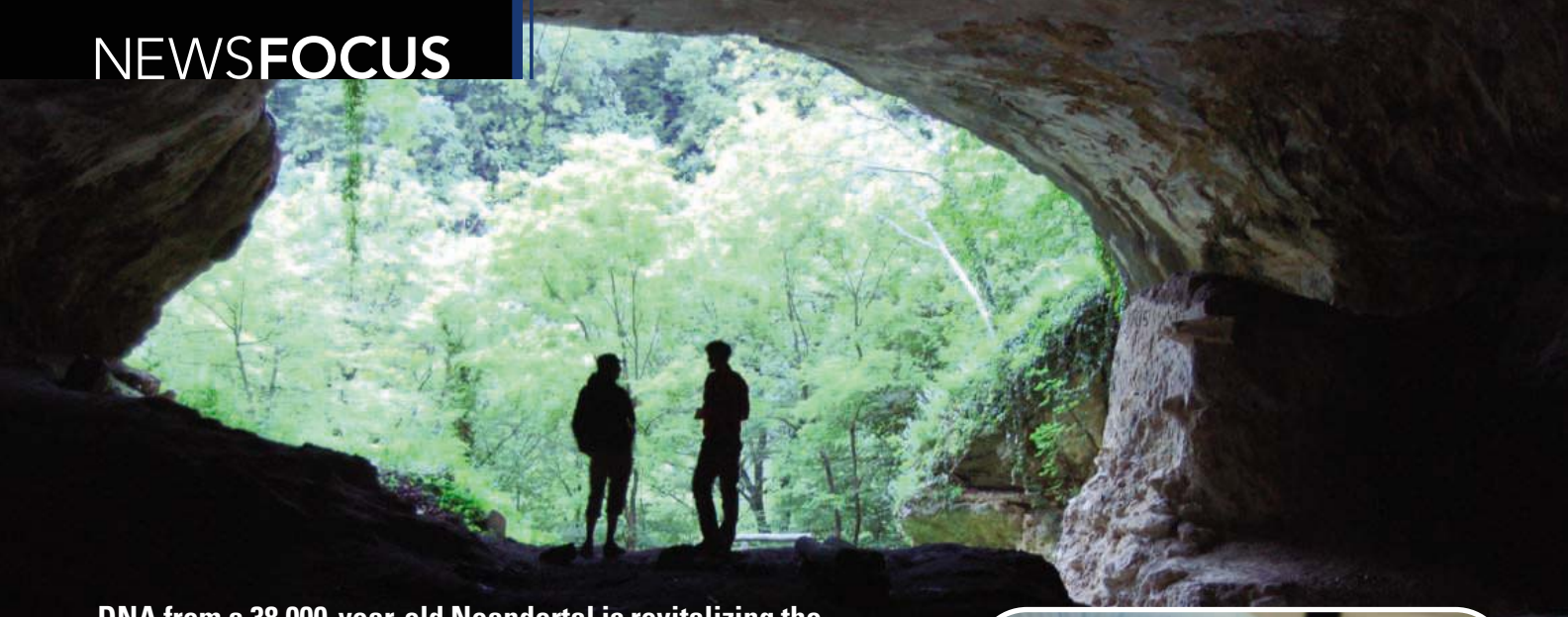
International desire for more cooperation from China played a key role in the final vote between Chan and Frenk, several observers say. Richard Horton, editor of *The Lancet*, who before the election made no secret of his support for Frenk's candidacy, says the result was based on political calculations rather than personal differences between the candidates. "The vote … was as much a vote for China as it was for Margaret Chan," he says.

Chan, 59, who was born in Hong Kong and lived there most of her life, studied medicine at the University of Western Ontario in Canada and public health at the National University of Singapore. In Hong Kong, she instituted a "diapers to grave" approach to public health, with a focus on preventative care and encouraging healthy lifestyles.

In explaining her priorities after her election, Chan said that the people of Africa "carry an enormous and disproportionate burden of ill health and premature death," and raising their status therefore must be one of the key measures of WHO's performance. Women's health is another key indicator, she said.

She emphasized that improving women's health means addressing not only reproductive health issues but also indoor air pollution from cooking fires, multiple infectious diseases, and violence. Targeting such problems improves the health of entire families and communities, she argued.

Even so, Horton predicts, her priorities could bring her into conflict with the United States, which campaigned hard for her election behind the scenes. "She can't deal with [women's health] without contraception, abortion, and condoms. … It's going to take her into deeply political territory, and that's good. That's what we need WHO to do," he says. "She has set out a clear agenda. It's a good agenda. Now we need to give her the benefit of the doubt." **–GRETCHEN VOGEL**



DNA from a 38,000-year-old Neandertal is revitalizing the once-moribund field of ancient DNA, and it promises a fresh perspective on how we differ from our closest relatives

The Dawn of Stone Age Genomics

WHEN GERMAN QUARRY WORKERS CHIPPED the first Neandertal bones out of a limestone cave in 1856, DNA analysis wasn't even a glimmer in any scientist's mind. Now, two reports, one on page 1113 and the other in the 16 November issue of *Nature*, describe the first successes in sequencing nuclear DNA from a Neandertal bone—a feat once considered impossible. The results from the two groups, working collaboratively but using different approaches, support the view that Neandertals are a separate branch of the hominid family tree that diverged from our own ancestors perhaps 450,000 years ago or more.

Because the extinct Neandertals are our closest relatives, comparing their DNA to ours may one day reveal the mutations that set *Homo sapiens* apart from all other species, as well as the timing of key evolutionary changes. But it's early days yet, and this week's papers chiefly suggest the potential of Neandertal genomics. They also fan the flames of the debate about how different Neandertals were from modern humans, and whether the two groups interbred during the thousands of years they coexisted in Eurasia (see sidebar, p. 1071). "This is great stuff," says molecular evolutionist Alan Cooper of the University of Adelaide, Australia. "It opens the way for much more work on identifying uniquely human genetic changes."

Coming on the heels of dramatic sequencing successes with ancient mammoth and cave bear DNA, the papers also herald a renaissance for a field that has been stymied by issues of poor sample quality and contamination. The Neandertal studies use metagenomics, which makes unnecessary the onerous task of purifying ancient DNA. They also employ faster, cheaper sequencing methods, and their achievement demonstrates the feasibility of deciphering ancient genetic material. "It has people talking about new ideas, new extraction techniques, new ways to prepare samples, new ways to think about old DNA," says Beth Shapiro, an ancient DNA specialist at the University of Oxford in the U.K.

Both teams are planning major additional projects. In July, the team led by Svante Pääbo, a paleogeneticist at the Max Planck Institute for Evolutionary Anthropology in Leipzig, Germany, announced that it plans to produce a very rough draft of the entire Neandertal genome in 2 years. With that draft, he and others will be better able to tell which of the 35 million bases that differ between chimp and humans are mutations that occurred in just the past 500,000 years and therefore likely define our species. "Perhaps we can find that last little bit that made us special," says Pääbo.

Meanwhile, the other team, led by Edward Rubin, head of the Department of Energy Joint Genome Institute in Walnut Creek, California, has support from the U.S. National Institutes of Health to gather DNA from several Neandertal fossils to study specific regions deemed key to understanding human evolution. At least one other team, led by Cooper, has its own Neandertal project and is working to gather DNA from other ancient humans as well. "A whole new world has opened up with regard to what can be done with ancient DNA," says Thomas Gilbert, a paleogeneticist at the University of Copenhagen, Denmark.

But despite the seductive promise of new techniques, researchers warn that ancient DNA has been a fickle mistress. Over the past 20 years, successes have been followed by frustration after frustration. It's hard to find suitable DNA, and it's also quite tricky to avoid contamination with modern genetic material and to cull errors. These issues may come back to haunt Pääbo and Rubin, says genomicist Stephan Schuster of Pennsylvania State University in State College. "The divergence [between living people and Neandertals] is so small compared to the DNA damage and the sequencing error" that

CREDITS: JOHANNES KRAUSE/MPI-EVA; (INSET) MPI-EVA

Rare find. Neandertal bone (*inset*) from this Croatian cave had well-preserved DNA, which has now been sequenced.

DNA-free. Clean-room garb in Spain's El Sidron cave helps reduce contamination by human DNA.

it's hard to be confident of any results, he says. "If we've learned anything, it is that we generally haven't perceived the full extent of the problems and complexities of ancient DNA research," admits Cooper. "We're still very much in the learning curve."

Ups and downs

Ancient DNA made its first appearance in 1984, when Allan Wilson of the University of California (UC), Berkeley, was able to tease out 100 bases from a quagga, an extinct species that looked like a cross between a horse and a zebra. A year later, Pääbo succeeded in extracting genetic material from a 2400-year-old Egyptian mummy.

The world was wowed by these successes, "but there was not much future in the field or the approach," Pääbo recalls. DNA degrades after death, as water, oxygen, and microbes attack it, and the sequencing methods of the time demanded more DNA than was readily available from ancient specimens.

The polymerase chain reaction (PCR), which uses an enzyme to make millions of copies of a particular DNA fragment, seemed to be just what the field needed, offering a way to amplify and read a tiny bit of sequence. The technique powered analyses of quagga, Tasmanian wolves, moas, and other extinct species during the 1990s.

But reliable results from more ancient specimens proved hard to come by. The reaction also amplified age-induced errors and extraneous DNA. A few spectacular failures cast doubt on the whole field: Supposedly 25-million-year-old DNA from amber-encased bees and even older DNA from dinosaurs turned out to be from living humans instead. Ancient human remains were especially problematic because of the specter of contamination: Anyone who handled bone could leave traces of their DNA upon it, and it was impossible to distinguish old from modern sequence.

Then in 1997, following new methodological guidelines, a team led by Pääbo, then at the University of Munich in Germany, and his student Matthias Krings restored the appeal of ancient DNA by decoding 379 bases of Neandertal mitochondrial DNA (mtDNA) (*Science*, 11 July 1997, p. 176). The bases were quite different from the equivalent modern human DNA, suggesting that Neandertals were a distinct species that split off from a common ancestor a half-million years ago and did not interbreed with modern humans. That and subsequent mtDNA and fossil studies supported the leading view that *H. sapiens* arose in Africa and spread around the globe, replacing other kinds of humans.

But in part because modern humans and Neandertals overlapped in Europe and west Asia for at least a few thousand years, and perhaps up to 10,000 years, some researchers had continued to argue that the two species interbred. They pointed out that 379 base pairs were too few to be conclusive. Also, because mitochondria are passed on only by the mother, nuclear DNA is needed to rule out the possibility of mixing.

Not quite gone. Genome data may one day shed light on how Neandertals lived.

Making the dream real

But getting nuclear DNA from ancient bones was a tall order. Back in 1997, "it was just a dream," Pääbo recalls. Because the amount of nuclear DNA in a cell is just 0.05% that of mitochondrial DNA, it's even harder to get enough nuclear DNA to sequence, particularly because often the DNA has disintegrated. Also, Neandertal bones are rare, and curators are reluctant to provide samples. But Pääbo's team devised a hierarchy of tests that required just a tiny amount of material to begin with.

First they tested a tiny, 10-milligram sample for intact proteins, as their presence suggests that DNA was preserved as well. Then they examined 150 milligrams to determine the ratio of Neandertal to modern human DNA, using existing Neandertal mtDNA as a guide. Two of the 70 samples they examined passed both tests with flying colors. So Pääbo's team sliced out a larger piece of one, a 38,000-year-old bone from Croatia, and extracted the DNA.

Meanwhile, Rubin had begun to think that the metagenomics approaches that he was pioneering to study microbial diversity would work with fossil DNA too. He suggested to Pääbo that Neandertal genomics might now be possible. After Rubin's postdoc James Noonan successfully sequenced 26,861 bases of cave bear DNA (*Science*, 22 July 2005, p. 597), Pääbo gave a sample of the Neandertal DNA to Noonan to work on.

The two teams embarked on parallel but independent analyses using different methods. Noonan first created a library of Neandertal DNA incorporated into live bacteria. As each bacterium replicated, it made copies of a particular fragment. The team employed a new, massively parallel technique called pyrosequencing, which uses pulses of light to read the sequence of thousands of bases at once. Sophisticated computer programs then compared the sequence fragments to available DNA databases and identified the potential Neandertal ones based on their similarity to modern human sequence. The team used several tests to rule out contamination with modern human DNA, such as checking that fragments had the correct flanking sequence and the expected amount of DNA damage for their size. In all, Rubin's team was able to extract 65,000 bases of Neandertal DNA.

CREDITS (TOP TO BOTTOM): EL SIDRON (ASTURIAS, SPAIN), PHOTO BY J. FORTEA; THE BRIDGEMAN ART LIBRARY/GETTY IMAGES

Pääbo employed pyrosequencing too, but he used a different method to prepare the DNA. Schuster and Hendrik Poinar of McMaster University in Hamilton, Canada, had successfully used this technique to read an astonishing 13 million bases from a 27,000-year-old mammoth (*Science*, 20 January, p. 392). This procedure avoids using bacteria, which for unknown reasons sometimes fail to incorporate certain stretches of DNA and so may not provide a complete sequence. Instead, Pääbo's team coated tiny beads with Neandertal DNA fragments, one fragment per bead. Then each bead's DNA was amplified, independently, by PCR, and read using pyrosequencing.

Ed Green of Pääbo's lab and his colleagues sequenced 225,000 fragments of DNA, totaling millions of bases. But by comparing the sequences with those in existing databases, they found that "the vast majority [of the DNA]—94%—has nothing to do with the human genome," says Pääbo, and came from sources such as soil microbes. Still, they identified a staggering 1 million bases of Neandertal DNA.

Green kept tabs on contamination in part by comparing stretches of mtDNA that showed up in the sequencing to known modern human and Neandertal mtDNA. They found little modern human mitochondrial sequence and say they are confident their Neandertal DNA is genuine.

Both teams compared the new sequences to the modern human genome and to the chimp genome and tallied the sequence differences between each pair of species. Places where the two human genomes match but the chimp's differs likely mark mutations that resulted in uniquely human changes, perhaps including our upright skeletons, bigger brains, lack of hair, and so forth. Differences between the two humans are signposts to changes that were key to their individual evolution. Eventually those changes could lead researchers to the genetic basis of *H. sapiens* speciation.

As expected, the Neandertal and human genomes proved more than 99.5% identical. Rubin's team's analysis of 65,000 bases revealed that the two humans shared 502 mutations that were different from chimp bases. And 27 bases varied between modern humans and Neandertals, indicating sites where evolution occurred after the two species diverged. Assuming that chimps and humans split 6.5 million years ago, the most recent common ancestor of the two human species lived 468,000 to 1 million years ago, most likely dating back 700,000 years, Noonan and his colleagues report.

In Green and Pääbo's much larger analysis, 10,167 bases were shared by just the modern human and Neandertal, and 434 were unique to modern humans. Taking a slightly different approach from Rubin, the Leipzig team found a more recent divergence time, about 465,000 to 569,000 years ago. This matches the mtDNA analyses, too, but doesn't quite settle the question. Not everyone agrees with the 6.5-million-year-old divergence date for humans and chimps, and a different date would change the timing of the split between modern humans and Neandertals.

As to the question of admixture, Rubin's group found no sign of it. There were no sites where the Neandertal possessed a rare single nucleotide polymorphism (SNP) found only in Europeans, which one would expect had interbreeding occurred. However, given the size of the study, there's still a chance that such shared SNPs exist but haven't yet been found, Rubin explains. So his study refutes the notion that Neandertals were major contributors to the modern human genome but can't rule out a modest amount of gene flow.

In contrast, the Leipzig group did find some evidence of hanky-panky between the two humans—although it's far from conclusive. They used the HapMap and another large catalog of modern human variation developed by a private company to guide them to potential SNP sites in the Neandertal. They found that at 30% of those sites, the Neandertal had the same base as living people, but the chimp had a different base. That's too much similarity, given how long ago the two lineages split. "Taken at face value, our data can be explained by gene flow from modern humans into Neandertals," says Pääbo. He thinks there may have been one-sided mating: Modern human males invaded the Neandertal gene pool by sometimes fathering children with Neandertal females, but not necessarily vice versa.

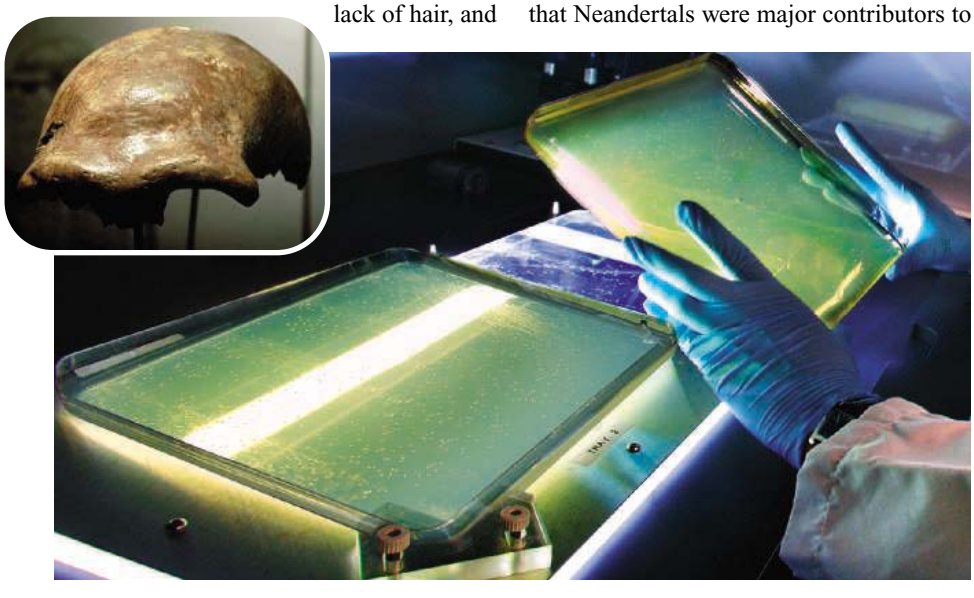
To those who have long argued for Neandertal admixture—and been in the minority—this is vindication. "These comprise some of the strongest genetic evidence of interbreeding with Neandertals that we have yet seen," says Milford Wolpoff, a biological anthropologist at the University of Michigan, Ann Arbor. But Stanford paleoanthropologist Richard Klein disagrees. "I don't think either paper bears much on the issue of admixture," he says. Schuster is even more circumspect: "Both papers are overinterpreting the data."

Rubin hopes that other researchers will do their own analyses on these publicly available data to help clarify the results. But Montgomery Slatkin, a theoretical population geneticist at UC Berkeley, thinks that even with more studies and more sequence, "it will be very difficult to distinguish between a low level of admixture and no admixture at all."

Concern about contamination

Anxiety about the sequence being wrong fuels this pessimism. Researchers need to be sure that what they called "Neandertal" isn't really "technician" DNA. And contamination is hard to avoid. "Bone acts like a sponge; a drop of sweat on the surface will penetrate very deep," Schuster explains.

With nonhuman ancient DNA, researchers can easily pick out and discard modern sequences, but that's not possible with Neandertal DNA, which is nearly identical to our own, notes paleogeneticist Carles Lalueza-Fox of the University of Barcelona, Spain. He is not convinced that the tests for



From bones to genomes. Plates of bacteria can reproduce DNA from bones like this skullcap (*inset*) from the Neander Valley.

CREDITS: ROY KALTSCHMIDT/LAWRENCE BERKELEY NATIONAL LABORATORY; (INSET) THE BRIDGEMAN ART LIBRARY/GETTY IMAGES

A Neandertal Legacy?

The perennial question about Neandertal-human relations is, "Did they mate?" (*Science*, 11 February 2005, p. 841). The lack of a strong Neandertal signature in the modern human genome means that such interspecies dalliances were probably rare, but the Neandertal nuclear DNA sequenced to date raises the possibility that interbreeding did happen (see main text). If so, there may be traces of Neandertal genes in living people, especially if the Neandertal variants were favored by natural selection. Now a handful of other studies are finding genes that may fit the bill.

"There is now a relatively long list of candidates" for such adaptive genetic variants, contends anthropologist John Hawks of the University of Wisconsin, Madison. But not all researchers agree. Population geneticist Laurent Excoffier of the University of Bern in Switzerland counters that it's "highly unlikely" there were enough matings between Neandertals and modern humans to have left significant traces in the modern genome.

The most recent candidate was reported last week in the *Proceedings of the National Academy of Sciences* by a team led by geneticist Bruce Lahn of the University of Chicago in Illinois. Lahn's team had earlier claimed that a variant of the brain-related gene *microcephalin* first appeared in modern humans about 37,000 years ago and quickly spread around the world because it was favored by selection (*Science*, 9 September 2005, p. 1662). In the new work, Lahn estimated that the variant actually arose in hominids more than 1 million years ago, long before it appeared in our own lineage. He suggests interbreeding, probably with Neandertals, as a likely explanation. "It seems to be the most compelling case to date for a genetic contribution of Neandertals to modern humans," says Svante Pääbo of the Max Planck Institute for Evolutionary Anthropology in Leipzig, Germany.

Kissing cousins? Neandertals (*foreground*) may have left some genes behind.

Similar candidates include a gene shown to have conferred a reproductive advantage in living Icelanders, a variant of a gene called *MAPT* implicated in neurological disease. As with *microcephalin*, the *MAPT* variant appeared in modern humans about 30,000 years ago but apparently arose in hominids much earlier and so may have come from Neandertals, according to recent work by John Hardy of the National Institute on Aging in Bethesda, Maryland.

There are several genetic variants whose roots go back as far as 2 million years ago but appeared more recently in modern humans, says geneticist Michael Hammer of the University of Arizona in Tucson. He says this pattern is best explained by occasional matings among different hominid groups within Africa as well as between African migrants and Eurasian hominids, including possibly *Homo erectus*. Even Chris Stringer of the Natural History Museum in London, who has argued that modern humans migrating from Africa replaced Neandertals with little or no interbreeding, now says that some interspecies matings are "feasible."

Just why genes from Neandertals or other ancient hominids would have benefited modern humans remains a mystery. But if the geneticists are correct, it could mean that before Neandertals went extinct about 30,000 years ago, they left modern humanity with lasting gifts.

–MICHAEL BALTER

contamination are foolproof. "It might never be possible to determine if the amplified sequence is real or one of the many potential sources of contamination," agrees Shapiro.

All the same, researchers are making some headway. Lalueza-Fox sequenced mtDNA from everyone who had ever touched a Neandertal specimen and compared it to the DNA obtained from the Neandertal. He found that most of the contamination came from the field, not the lab. His solution: Treat the excavation site like a crime scene. Archaeologists in his team now wear face masks, coveralls, and sterile gloves; they use sterile blades and quickly freeze bones destined for DNA sampling. The dress code has reduced human contamination from about 95% to 5%, says Lalueza-Fox.

Even if contamination can be contained, ancient DNA studies must contend with errors. Sequencing itself makes mistakes. And that's where Rubin's bacterial libraries come in handy. With an ever-reproducing source of DNA, his team can sequence the same fragment multiple times and therefore tell right from wrong bases. With Pääbo's method, the sample gets used up.

More problematic are those errors that have arisen from age-related decay. "Many, and perhaps most, observed differences between a Neandertal genome sequence and the human reference will be caused by [ancient] chemical damage to the Neandertal sample," says Webb Miller, a computer scientist at Pennsylvania State University. One way to detect such errors is to sequence and compare several different specimens, because each fossil should have a unique pattern of DNA damage, says Miller.

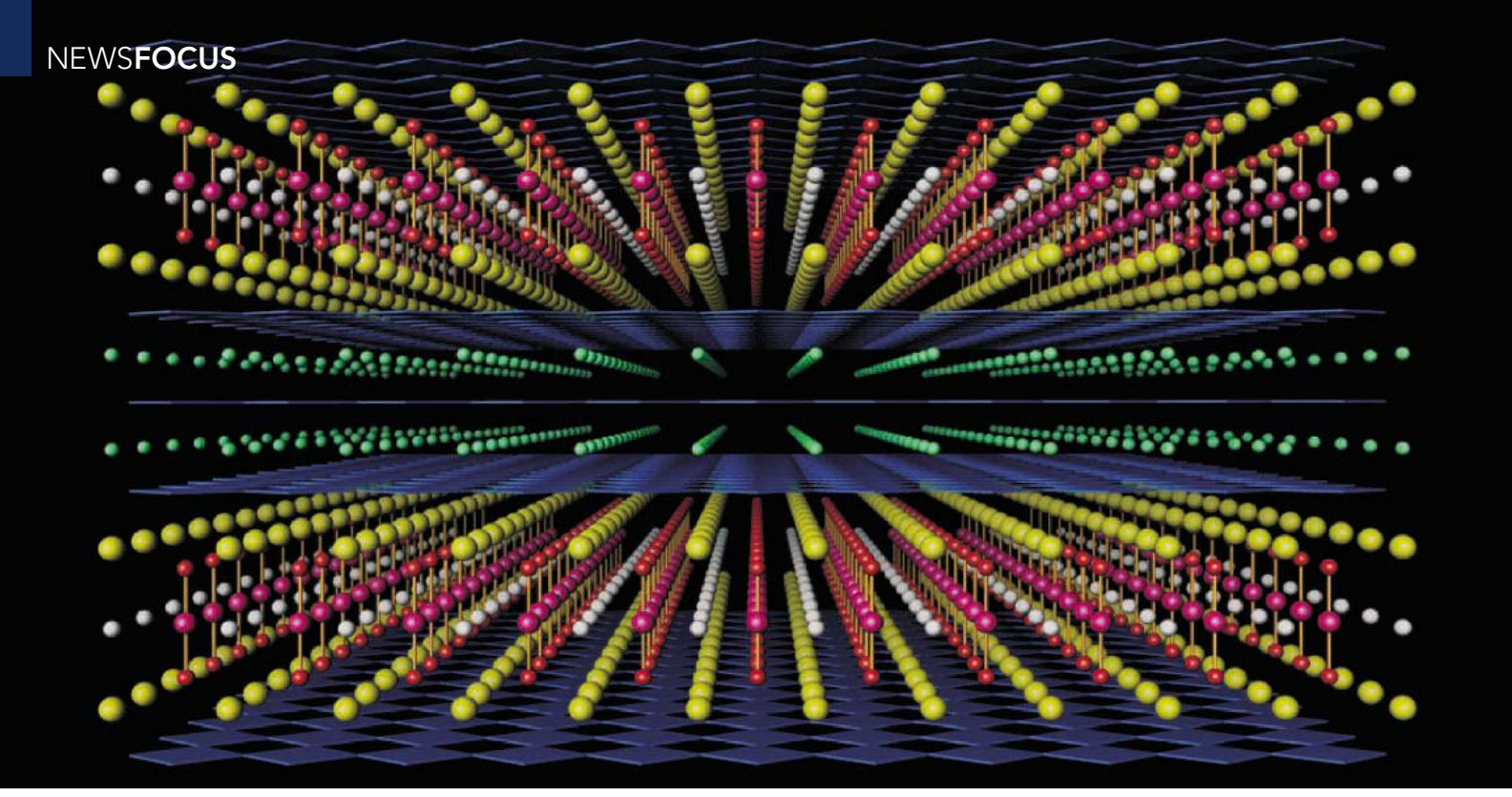
Here, too, Rubin's methods can help. He envisions several libraries, each from a different Neandertal. Researchers would pull out the same fragment from each library to compare with each other and with living people. A pilot project has already demonstrated probes that ferret out specific target sequences, so the team needn't analyze the billions of bases shared by Neandertals and living humans, or among different Neandertals. "We will be able to identify and confirm sequence changes in more than one Neandertal without having to sequence several Neandertals to completion," Rubin says. "Seeing the same change in multiple Neandertals will give us confidence that we got [the sequence] right."

Such talk of multiple sequencing has some fossil guardians anxious. "If everybody that wanted a chunk of Neandertal got a chunk of Neandertal, that would put the whole Neandertal fossil record at risk," warns paleoanthropologist Tim White of UC Berkeley.

At this point, however, even the paleontologists seem eager to see what genomic studies can do. This month, Lalueza-Fox will bring one of his "clean-room excavated" bones to Pääbo to see whether its DNA qualifies for sequencing, and he's thrilled with the potential of sequencing. "For the [150th] Neandertal anniversary, we are moving from paleogenetics to paleogenomics," Lalueza-Fox explains. "It is incredible considering this was impossible just a few years ago."

–ELIZABETH PENNISI

CREDIT: FRANK FRANKLIN II/AP PHOTO



After 2 decades of monumental effort, physicists still cannot explain high-temperature superconductivity. But they may have identified the puzzles they have yet to solve

High T_c: The Mystery That Defies Solution

TWENTY YEARS AGO, A FIRESTORM OF discovery swept through the world of physics. German experimenter J. Georg Bednorz and his Swiss colleague Karl Alexander Müller kindled the flames in September 1986 when they reported that an odd ceramic called lanthanum barium copper oxide carried electricity without any resistance at a temperature of 35 kelvin—12 degrees above the previous record for a superconductor. The blaze ran wild a few months later when Paul Chu of the University of Houston, Texas, and colleagues synthesized yttrium barium copper oxide, a compound that lost resistance at an unthinkable 93 K—conveniently warmer than liquefied air.

A frenzy of slapdash experimenting and sensational claims ensued, says Neil Ashcroft, a theorist at Cornell University. He organized a session on the new high-temperature superconductors at the meeting of the American Physical Society in New York City the following March. The "Woodstock of physics" stretched until 4 a.m. and bubbled over with giddy enthusiasm. "We had prominent people saying it would all be explained quickly and that we would have superconducting power lines and levitating trains," Ashcroft says.

Ashcroft himself had doubts, however, as he told a class of graduate students a few months later. (I was a member of the class.) The materials comprised four and five elements and possessed elaborate layer-cake structures. They broke the rules about what should make a good superconductor. In short, Ashcroft predicted, high-temperature superconductivity would remain the outstanding problem in condensed matter physics for the next 25 years.

That prognostication is coming true. Two decades after high-temperature superconductors were discovered, physicists still do not agree on how electrons within them pair to glide through the materials effortlessly at temperatures as high as 138 K. Researchers haven't failed for lack of trying. According to some estimates, they have published more than 100,000 papers on the materials. Several theorists claim they have deciphered them—although their explanations clash. Still, high-temperature superconductivity has refused to submit to some of the world's best minds.

"The theoretical problem is so hard that there isn't an obvious criterion for *right*," says Steven Kivelson, a theorist at Stanford University in Palo Alto, California. Experimenters are producing a flood of highly detailed data, but physicists struggle to piece the results together, says Joseph Orenstein, an experimenter at the University of California, Berkeley, and Lawrence Berkeley National Laboratory. "It must be close to unique to have so much information and so little consensus on what the questions should be," Orenstein says.

The problem is more than a sliver under the nail. High-temperature superconductivity has shown that physicists' conceptual tools can't handle materials in which electrons shove one another so intensely that it's impossible to disentangle the motion of one from that of the others. Such "strongly correlated" electrons pop up in nanodevices and novel magnets, organic conductors and other exotic superconductors. "High-temperature superconductivity is the stumbling block of the whole discipline of condensed matter physics," says Peter Abbamonte, an experimenter at the University of Illinois, Urbana-Champaign.

In spite of the difficulty of the puzzle, many physicists say they are closing in on a solution. Most now agree on certain key properties of the materials. Precision experiments are

CREDITS (TOP TO BOTTOM): P. EDWARDS AND M. O. JONES/UNIVERSITY OF OXFORD

Hot stuff. The structure of mercury barium calcium copper oxide, a superconductor at 138 K.

revealing surprising details of the compounds. And computer simulations—and perhaps even mockups fashioned of ultracold atoms and laser light—could soon show physicists whether their basic model of the problem is correct. "If I had to make a prediction," Kivelson says, "I would say that in 10 years time the problem will be solved."

The ultimate chess game

Even "conventional" superconductivity, which was discovered in 1911, is mind-bending. Electrons in a metal move in quantum waves of distinct energies. Quantum mechanics prohibits two electrons from occupying the same wave or "state," so they stack into the states from the lowest energy on up. But when metals such as lead and niobium are cooled to near absolute zero, the electrons in them can lower their total energy by pairing like ballroom dancers. That partnership produces superconductivity, as explained in 1957 by theorists John Bardeen, Leon Cooper, and John Robert Schrieffer.

The pairing alters the spacing of the rungs on the energy ladder, creating a gap near the top of the stack. To break from its partner, an electron must jump the gap to an empty state. There isn't enough energy around to allow that, so the pairs glide along unperturbed. Something must glue the pairs together, and according to the Bardeen-Cooper-Schrieffer (BCS) theory, the adhesive is quantized vibrations of the crystalline material, or "phonons." A passing electron attracts the slower-moving ions in the crystal lattice, which squeeze together to produce a knot of positive charge that attracts another electron (see diagram).

High-temperature materials literally take superconductivity to a new plane. The compounds contain planes of copper and oxygen ions that resemble chess boards, with a copper ion at every corner of a square and an oxygen ion along each side. Electrons hop from copper ion to copper ion. Between the planes lie elements such as lanthanum, strontium, yttrium, bismuth, and thalium. But it is along the copper-and-oxygen planes that the electrons pair and glide.

Just how that happens is anything but clear. The electrons in an ordinary metal hardly notice one another and interact mainly with phonons. In contrast, the electrons in high-temperature superconductors shove one another so mightily that they tend to jam up with one electron on each copper ion, like gridlocked commuters. That impasse can be broken only by tweaking the material's chemical composition to siphon away some of the electrons to create positively charged "holes," a process called doping.

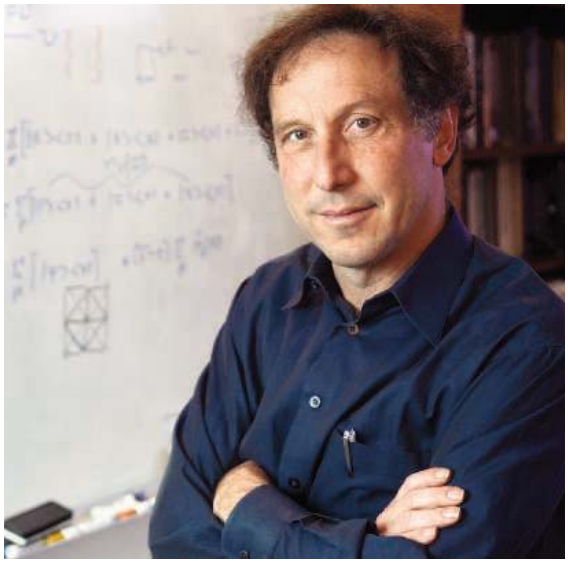
The challenge then is to explain how electrons that fiercely repel each other manage to pair anyway. Some researchers argue that waves of magnetism play a similar role to the one phonons play in conventional superconductors. Others focus solely on how the electrons shuffle past one another in a quantum-mechanical game of chess. Still others say that patterns of charge or current, or even phonons, play a crucial role. Pairing might even require all of these things in combination, which would be many physicists' nightmare scenario.

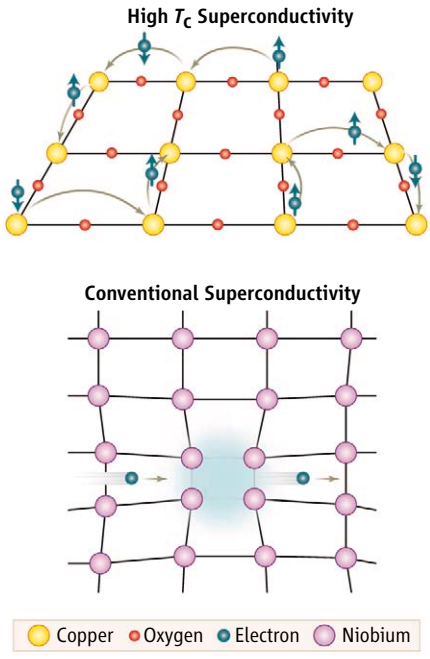
Familiar solutions

Some of the theories being refined today emerged soon after Bednorz and Müller's discovery, and the dividing lines that run through the field were drawn in those heady days. For example, as early as 1987 some theorists argued that high-temperature superconductivity arose not from phonons but from the interaction of the electrons alone. But even those who agree on that principle often disagree on the details.

The idea that waves of magnetism drive the superconductivity is based on the fact that electrons act like little magnets. Those on adjacent copper ions point in opposite directions, creating an up-down-up-down pattern known as antiferromagnetism. The electrons can tilt and flip, and waves of wobble coursing through this arrangement can provide the glue for pairing, says David Pines, a theorist at Los Alamos National Laboratory in New Mexico and the University of California, Davis.

"The theoretical problem is so hard that **there isn't an obvious criterion for *right*.**"
—Steven Kivelson, Stanford University





Shall we dance? Instead of the motion of ions, the subtle waltz of electrons along atomic planes may cause pairing in high-temperature materials.

But Philip Anderson, a theorist at Princeton University, says that no glue is necessary. Just months after the discovery of high-temperature superconductors, he proposed a scheme known as the resonating valence bond (RVB) theory, which focuses on subtle quantum connections between electrons on neighboring copper ions. In the theory, no waves of any kind pass between electrons, Anderson says.

Thanks to the weird rules of quantum mechanics, each electron can point both up and down simultaneously. Moreover, neighboring electrons can join in an odd quantum state called a singlet in which both electrons point both up and down at once, but the two electrons always point in opposite directions—either down-up or up-down. When enough holes open in the plane, singlets form and begin to slide freely past one another, eventually producing superconductivity.

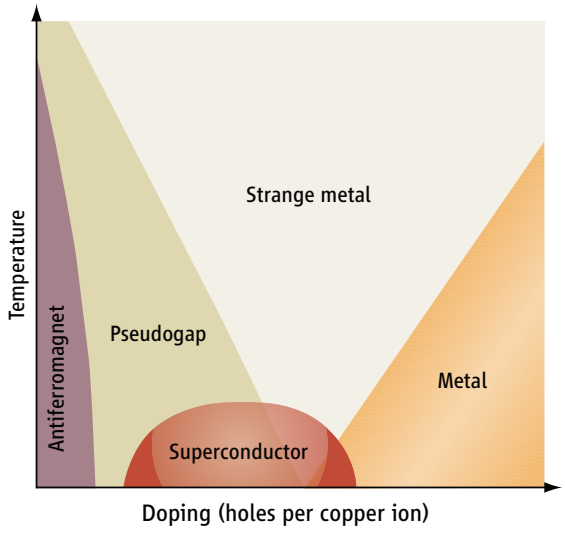
Others contend that both the magnetic fluctuation and RVB theories leave out some essential piece of physics. Stanford's Kivelson believes stripes of electric charge on the planes, which have been seen in some materials, may be necessary to trigger the pairing. Chandra Varma, a theorist at the University of California,

CREDITS (TOP TO BOTTOM): P. HUEY/*SCIENCE*; LINDA A. CICERO/STANFORD NEWS SERVICE

Riverside, proposes that loops of current flowing inside each copper-and-oxygen square are key.

Still others argue that high-temperature superconductivity may not have one root cause. "There is no silver bullet that is going to explain everything," says Thomas Devereaux, a theorist at the University of Waterloo in Canada. The fact that materials with very similar structures have very different critical temperatures shows that the copper-and-oxygen planes are not the whole story, he says. Phonons may still play an essential role, such as driving up the critical temperature, Devereaux says.

As in the beginning, the field is contentious. Recent experiments have hinted at current loops. "If these are accepted, the theoretical game is over," Varma says. "That's why no one wants to accept it." Anderson is equally convinced that his RVB theory is correct—and underappreciated. "Eighty percent of the field is against anything—especially anything that might solve the problem," he says.



Terra incognita. The mysterious and controversial pseudogap phase may hold the key to explaining superconductivity.

Mapping out the mysteries

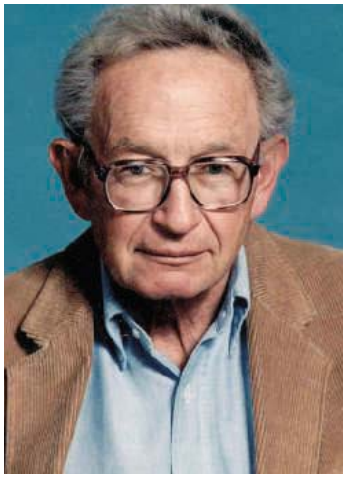
In spite of the discord, researchers have made progress—especially the experimenters. For example, in 1994, John Kirtley and Chang Tsuei of IBM's T. J. Watson Research Center in Yorktown Heights, New York, probed the shape of the cloudlike quantum wave that describes the paired electrons. In a conventional superconductor, electrons can pair in any direction and can sit on top of each other, so the wave is a sphere. In high-temperature superconductors, Kirtley and Tsuei found, the cloud is shaped like a four-leaf clover. That "d-wave" shape means that paired electrons sit on adjacent copper ions and never on the same ion.

D-wave pairing would be hard to explain with phonons, but it had been predicted by Anderson and others who favored purely electronic theories. As a result, even most of those who say phonons play a role do not believe that they alone cause pairing.

By dint of a variety of experiments, researchers have also agreed upon the properties common to all the materials, which change with the amount of doping. Cook up an undoped material, and it's an antiferromagnetic insulator. Dope it to draw between 6% and 22% of the electrons out of the planes, and it's a superconductor. Dope it more, and it becomes an ordinary metal.

These properties can be plotted on a "phase diagram" that, like some medieval map, charts the mysteries physicists face (see figure, above). "To solve the whole problem, you're going to need to understand the whole phase diagram," says Séamus Davis, an experimenter at Cornell University. "It could be that focusing on the mechanism is the reason that the mechanism hasn't been found."

Most intriguingly, at low doping a gap opens in the ladder of electron energy levels even at temperatures far above the superconducting transition. That "pseudogap" suggests that electrons pair at such toasty temperatures, and that superconductivity arises when the "preformed" pairs gather into a single quantum wave, some researchers say. "Everything we have seen goes in that direction," says Øystein Fischer, an experimenter at the University of Geneva, Switzerland. And Tonica Valla of Brookhaven National Laboratory in Upton, New York, and colleagues present data online in *Science* this week (www.sciencemag.org/cgi/content/abstract/1134742) consistent with this interpretation.



"Eighty percent of the field is against anything—especially anything that might solve the problem."

—Philip W. Anderson, Princeton University

Preformed pairs are too much to swallow for other researchers, who say the pseudogap is a sign of something else that clashes with superconductivity. For example, Zhi-Xun Shen of Stanford University and colleagues argue online in *Science* this week (www.sciencemag.org/cgi/content/abstract/1133411) that there may be two different gaps. Either way, the strange state might hold the key to explaining high-temperature superconductivity, says Michael Norman, a theorist at Argonne National Laboratory in Illinois. "The thing that explains the pseudogap may explain the superconductivity as well," he says.

Computers and cold atoms

Ultimately, the mystery of high-temperature superconductivity may be solved not in the lab or at the theorist's chalkboard but in the heart of a computer. Some theorists have turned to numerical simulations of the electrons hopping around the copper planes. If everything springs from the interactions between the electrons alone, then all the different phases and perhaps even the pairing mechanism should emerge from such simulations, much as the double helix, genes, and the mechanism of transcription arise from chemical interactions between the building blocks of DNA.

The mathematics can vary, but theorists generally study a scheme known as the Hubbard model, in which the only adjustable parameters are the ease with which the electrons hop and the strength with which they repel each other. Tracking electrons on a grid might sound easy, but the complexity of the quantum-mechanical calculations limits researchers to grids of a few dozen lattice sites. And still they must use approximation schemes to keep the calculation manageable.

Such simulations have begun to reproduce pairing, stripes, and features of the pseudogap, says André-Marie Tremblay, a theorist at the University of Sherbrooke in Canada. Unfortunately, different approximation methods can lead to different results for the same parameters, says Douglas Scalapino of the University of California, Santa Barbara. But that's not necessarily a bad thing, he says, because that very sensitivity suggests that the Hubbard model can produce a variety of effects with only a little tweaking, just as high-temperature superconductors do. "I interpret that to mean we have the right model," he says.

Meanwhile, a wild new kind of simulation could be gearing up to

CREDIT (BOTTOM): BOB MATTHEWS

leave computer simulations in the dust. Physicists have begun to construct artificial crystals by suspending ultracold atoms in corrugated patterns of laser light. In such an "optical lattice," the spots of light play the role of the ions, and the atoms play the role of the hopping electrons. The setup might be used to create an incarnation of the Hubbard model with hundreds of lattice sites and parameters that researchers can tune just by adjusting the spacing and the brightness of the spots.

Several groups are already racing to produce such systems. "In very quick succession, we have jumped over the first few hurdles, and maybe the number of hurdles ahead of us is not that much bigger than the number behind us," says Wolfgang Ketterle, an experimenter at the Massachusetts Institute of Technology in Cambridge. Using optical lattices, experimenters could map out the phase diagram of the Hubbard model within a few years, says Henk Stoof, a theorist at Utrecht University in the Netherlands. "They have all the things they need to do it," he says.

"It could be that **focusing on the mechanism is the reason that the mechanism hasn't been found.**"
—Séamus Davis, Cornell University

But even if such simulations do produce superconductivity, they may not yield a conceptual understanding of the pairing, some researchers say. Others question the relevance of the simulations to high-temperature superconductors. "We don't know that the Hubbard model is what's going on in the [materials]," says Cornell's Davis. "That's a hypothesis."

A threshold

Even without a theory to explain the materials, physicists agree that the pursuit of high-temperature superconductivity has already paid off handsomely. "It has led to the discovery of new materials, of new states of matter, of new concepts," says Aharon Kapitulnik, an experimenter at Stanford University. Shen says that in their quest to unravel the phenomenon, experimenters have honed their techniques to new levels of sensitivity, precision, and speed. "High-temperature superconductivity has completely changed the landscape of experimental condensed matter physics," he says.

At the same time, condensed matter researchers have come to see high-temperature superconductivity as the gateway to a new area of study: strongly correlated electrons. "This problem of strongly correlated electrons is the new frontier," says Argonne's Norman, "and high-temperature superconductors have brought it to the fore." Two decades after their discovery, high-temperature superconductors are viewed less as a singular mystery and more as a threshold to new realms of physics. Physicists hope it won't take another 20 years to cross it. **–ADRIAN CHO**

APPLICATIONS

The Next Big Hurdle: Economics

Researchers have solved most of the technical challenges. Now companies are struggling to make HTS devices competitive

Six months after J. Georg Bednorz and Karl Alexander Müller discovered that a family of ceramics could conduct electricity without the electrical equivalent of friction, the scientific buzz swelled into full-scale hype. News accounts gushed at the prospect of magnetically levitated trains, novel sensors, superfast superconducting computers, and of course, lossless electricity transmission cables. For a generation that grew up watching the technological utopia of the Jetsons, the future, it seemed, was just around the corner.

The trouble is, it's still there. Two decades into the revolution, the effort to commercialize high-temperature superconductors (HTS) is not for the faint-hearted. Successful applications exist, although with names and roles that few people would recognize, such as current leads and cellular base station filters. And although those and other niche applications are turning a profit for their owners, the field is nothing like its founders envisioned. "In my opinion, we oversold high-temperature superconductivity," says Lucio Rossi, who heads the magnets and superconductors group at CERN, the European particle physics laboratory near Geneva, Switzerland.

Today's outlook is decidedly less rosy. "It's very difficult to make money on HTS," says John Rowell, a physicist at Arizona State University in Tempe, who notes that no venture capital–funded HTS company in the United States has ever had a year of profitability. Still, hope springs eternal, and after 20 years of development, HTS equipment makers seem to be finding ground beneath their feet. "It's a slow process," says Al Zeller, a superconducting magnet expert at Michigan State University in East Lansing. "But the applications are taking off." "The materials science in HTS has been terrific," says Bruce Strauss, who helps run the superconductivity program at the U.S. Department of Energy (DOE). "The engineering is just beginning. I've been seeing a lot more engineering than before of motors, coils, and so on. That's a good sign."

Slowing to a crawl

Part of what made the HTS revolution so exciting was that the novel superconductors looked and acted so differently from conventional low-temperature superconductors (LTS). The earlier materials were ductile metals, such as the alloy niobium-tin, that could be forged into wires for power cables or wound into spools for use in magnets, a key component for motors and generators.

HTS materials, by contrast, are brittle ceramics. In the early discoveries of HTS materials, researchers placed electrodes on opposite sides of a millimeter-sized ceramic fleck or perhaps a few-centimeters-long film of the material. That setup worked to show the drop in resistance characteristic of the onset of superconductivity. But nobody knew how to turn these hard, brittle flecks into kilometers of wire.

Part of the problem is that electrons passing through HTS materials, unlike those in conventional superconductors, prefer to travel in particular directions through the

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material's atomic lattice. Separate grains of the material must line up so that electrons can hop from one to the next; if the alignment between the atomic lattices of separate grains is off by more than several degrees, the conductivity of the material drops dramatically.

Researchers found that creating this alignment was fairly easy in a superconductor made from bismuth strontium calcium copper oxide (BSCCO). In BSCCO, the grains resemble flat rectangular plates. By 1988, researchers learned that by packing BSCCO in a silver tube and stretching the silver into a long thin wire, they could align the BSCCO grains closely enough to carry moderate supercurrents through the material.

Timeline of HTS Research and Development

2006
350-meter-long 2G wire carries >200 A HTS cable installed in Albany, New York, grid.

2003
5-megawatt HTS ship motor built and tested.

New spin. Compact HTS motors save fuel and space.

2002
Electronic structure of BSCCO shown to be disordered.

2000
1000-horsepower HTS motor built and tested.

1999
Meter-long 2G YBCO wire carries 122 A.

1996
50-meter HTS underground power cable built.

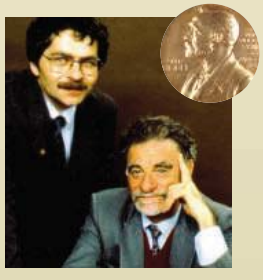
1995
First generation-2 YBCO wire.

Stripes of charge in high-T_c materials observed for the first time.

1994
1-km BSCCO wire.

IBM tricrystal experiment reveals d-wave symmetry.

1993
Thallium-doped mercury barium calcium copper oxide; T_c 138 K.



1989
First generation-1 BSCCO/silver HTS wire.

Thallium barium calcium copper oxide; T_c 125 K.

Experiments spot "pseudogap," possible sign of electron pairing above T_c.

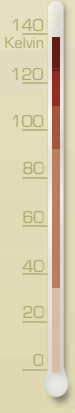
1987
Yttrium barium copper oxide (YBCO) discovered; T_c 93 K.

"Woodstock of Physics."

Bednorz and Müller share Nobel Prize in physics.

1988
Bismuth strontium calcium copper oxide (BSCCO); T_c 110 K.

1986
Lanthanum barium copper oxide discovered by Bednorz and Müller with T_c of 35 K.



Today, BSCCO wire technology has advanced considerably beyond its early days. Several superconducting wire companies now turn out kilometers of such first-generation (1G) wire. That has led to a slew of prototype devices, such as power cables, high-efficiency industrial motors, lightweight ship propulsion systems, and electricity-storing flywheels.

BSCCO has key shortcomings, however. Its silver sheathing makes it expensive, and strong magnetic fields sap its ability to carry a supercurrent unless the temperature around it drops much closer to absolute zero. But lowering the temperature limits its advantage over LTS materials.

In some cases, marrying 1G wires with more conventional materials can solve the magnetic-field problem. This year, for example, researchers at Sumitomo Electric in Japan unveiled a prototype motor for ship propulsion that relies on 1G wire wrapped around an iron core, which helps the wire withstand the high magnetic fields generated. The motor, a mere 10% of the volume and 20% of the weight of a conventional motor, also saves considerable fuel. A separate 36.5-megawatt HTS ship motor, produced by American Superconductor Corp. in Westborough, Massachusetts, has passed factory tests and is expected to be delivered to the U.S. Navy later this year.

Researchers around the globe have been making steady progress on another track as well. In 1995, physicists at Los Alamos National Laboratory in New Mexico and Oak Ridge National Laboratory in Tennessee turned a different cuprate superconductor—YBCO—into a 2.5-centimeter wire that could carry 1 million amps per square centimeter of cross section of the wire. Thanks to YBCO's cheaper starting materials and better ability to withstand high magnetic fields, the novel second-generation (2G) wire held out new hope for making cheaper magnetic field–resistant wires.

But scaling up those short wires turned out to be another major challenge for the field. YBCO's various-shaped grains proved much harder to orient than the platelike grains in BSCCO. Coaxing them into alignment required years of development work. Today, researchers and companies use a vari-

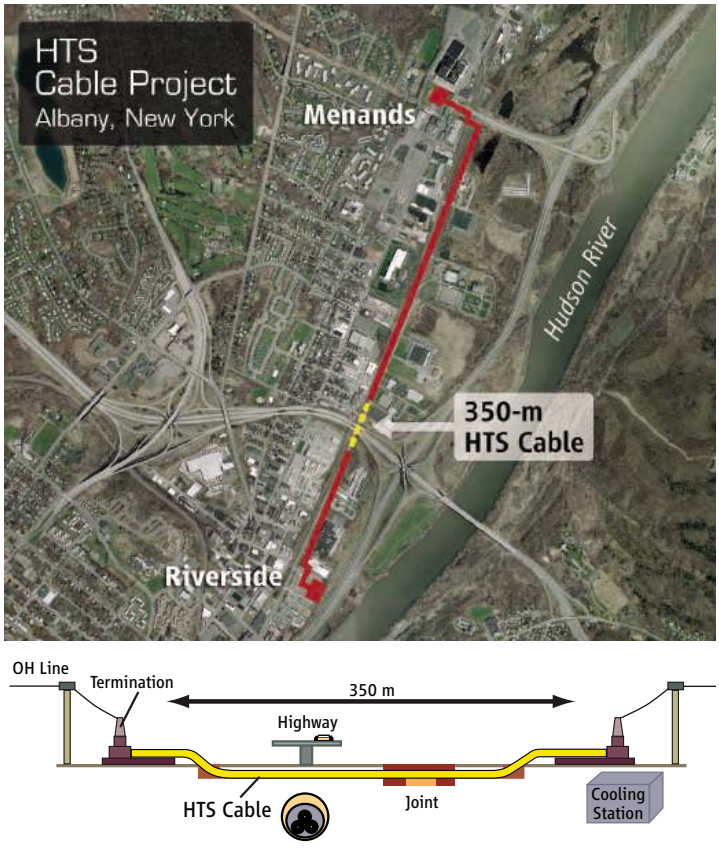
Time change. The discovery of new ceramic superconductors dominated the early years of HTS research. Today, the focus has migrated to applications.

CREDITS (TOP TO BOTTOM): SUMITOMO ELECTRIC INDUSTRIES LTD.; AMERICAN SUPERCONDUCTOR; IBM ZURICH RESEARCH LABORATORY; DATA SOURCE: DOE

ety of techniques to coat a nickel wire with ultrathin layers of materials that orient the YBCO grains as they grow on top.

Ripe for a change?

That strategy seems to be paying off. The August Applied Superconductivity Conference in Seattle, Washington, showcased several companies that are moving YBCO into applications. Xuming Xiong, a materials scientist with SuperPower, an HTS wire maker based in New York, reported that his company now routinely produces 700- to 800-meter-long 2G wires and is building a pilot plant to make wires that exceed 1 kilometer. Companies are already using these long 2G wires to make solenoid coils for superconducting magnets. Researchers at American Superconductor, for example, reported that they had made a solenoid coil that generates a 1.5-tesla magnetic field from 420 meters of superconducting coil. Researchers at Fujikura in Japan announced a similar 1-tesla magnet.



Fired up. An underground HTS power cable now connects two substations in Albany, New York. Such cables have the potential to carry three to five times more power than conventional copper cables and transmit it with higher efficiency.

With 1G wires now well into their commercialization effort and 2G wires working their way into prototype products, widespread applications of HTS now seem possible for the first time. The timing is fortunate, says James Daley, a superconductivity program manager at DOE, because the U.S. electricity grid is aging and due to be replaced, while demand for electricity continues to rise at about 2.6% per year. In many cases, utility companies can meet that rising demand by stringing extra power lines. But in many urban centers, gaining new rights of way to string power lines isn't possible. That has opened a window for superconducting cables, which can carry up to five times more power than conventional power lines can. DOE estimates that some 3500 kilometers of underground power cables in the United States could be converted to HTS cables.

Three demonstration cable projects are under way in the United States. On 20 July, for example, a collaboration of HTS companies, utility-grid operators, and a cooling specialty company connected the first HTS power cable to the U.S. grid in Albany, New York. That 350-meter cable is expected to supplement the current grid to carry enough extra energy to power more than 70,000 homes. Later this year, a 30-meter-long section of this HTS cable will be upgraded with a 2G cable.

Wires and cables are by no means the only starting materials for HTS applications. Researchers around the globe routinely craft dozens of different types of electronic, magnetic, and optical sensors from highly pure thin films of HTS materials. These sensors are used in applications as diverse as quantum computing experiments and medical imaging devices that track minute magnetic differences in tissue. For now, however, many experts think the biggest potential for HTS lies in the electric utility business. "The major impacts of HTS will not be felt until power applications become a reality," says Paul Chu, a superconductivity expert at the University of Houston in Texas and president of the Hong Kong University of Science and Technology.

Risky business

Despite the initial success of the power cables that have been installed so far, the HTS industry still faces an uphill battle. In the minds of utility executives, HTS "is still a very high-risk technology," says Alan Lauder, executive director of the Coalition for the Commercial Application of Superconductors in Kennett Square, Pennsylvania. Utility companies face a powerful disincentive to adopting new technology because they typically cannot pass the cost along to customers. They also worry about the reliability of unproven technology. "If you want something that will live for 30 years and never fail, you will not accept a 2-year demonstration project," says Lauder.

Some HTS applications are unique, however, or boast advantages that far outstrip those of conventional technology. One example, Lauder says, is utility-grid devices called fault current limiters that prevent power surges from destabilizing local grids. Another set of utility devices, called HTS dynamic synchronous condensers, help the grid maintain a near-uniform level of electrical "pressure" or voltage. Both applications are expected to be widely adopted in the next few years, Lauder and others say.

Still, despite a few successes, the outlook for HTS products remains mixed. Prototype magnetically levitated trains are running in Germany, China, and Japan, but it's not clear whether their use will spread. Plans for superconducting computers washed out years ago. And even one successful application—electronic filters used in the base stations that route cell phone calls—could be overtaken by improvements in conventional filter technology. "Customers do not care about science," says Arizona State's Rowell. "They care about a box with high performance and low cost."

Cost remains the biggest challenge for HTS devices, particularly for HTS wire that's priced three to five times higher than its copper equivalent. With the production of 2G wire now being scaled up, companies say they hope to close that gap by the end of the decade. But if the last 20 years have offered any lesson for entrepreneurs, it's that HTS hype inevitably yields to a more sober assessment of what the new science can deliver to the marketplace. **–ROBERT F. SERVICE**

Midnight oil. Müller (*left*) and Bednorz had to work nights to get access to equipment.

CREDIT: IBM ZURICH RESEARCH LABORATORY

PROFILE: KARL ALEXANDER MÜLLER AND J. GEORG BEDNORZ

Determined Duo Scored a Victory For Small-Scale Science

Two-man teams rarely win physics Nobels, but the discoverers of high-temperature superconductivity showed the power of in-depth knowledge and a good hunch

In 1986, the 75th anniversary of the discovery of superconductivity, the field was literally stuck in a deep freeze. Researchers' main goal was to raise the critical temperature (T_c) below which a material conducts electricity with no resistance, but progress had been slow. Starting at 4.2 kelvin in 1911 with mercury, T_c was pushed up from the 1930s onward with a series of intermetallic compounds—crystals made from different metals—all involving niobium. By 1973, the best T_c had reached just 23.3 K (in Nb_3Ge), and there it had stuck.

Researchers at IBM's Zurich Research Laboratory in Rüschlikon, Switzerland, like many others, decided it was time to take a new approach. In 1983, physicist Karl Alexander Müller, a Rüschlikon researcher for 2 decades, asked J. Georg Bednorz, a crystallographer specializing in materials known as perovskites who had joined IBM the previous year, to help him with an unlikely project: searching for new superconductors in complex metal oxides, materials usually known as insulators.

Müller had recently returned from a 2-year sabbatical at IBM's research center at Yorktown Heights, New York, and was fired with enthusiasm to study superconductors. Years of work with a perovskite oxide of strontium and titanium ($SrTiO_3$) had convinced him that perovskites had potential as superconductors. $SrTiO_3$ was already known to superconduct at the low temperature of 0.3 K, and in 1978, Bednorz, then at the Swiss Federal Institute of Technology (ETH) in Zurich, had collaborated with Rüschlikon's Gerd Binnig in coaxing the T_c of $SrTiO_3$ up to 1.2 K by doping the crystal with niobium. Another perovskite, made from barium, lead, and bismuth, had been shown in 1975 to have a T_c of 13 K.

Perovskite crystals with the right combination of metal ions, Müller concluded, would be conductors with a strong coupling between electrons and phonons, ripples in the crystal lattice that in conventional superconductors act as the glue to stick electrons together in pairs—an essential part of the superconducting process.

Bednorz and Müller started out with perovskites of lanthanum, nickel, and oxygen and systematically replaced varying amounts of the nickel with aluminium, the lanthanum with yttrium, and finally the nickel with copper. But superconductivity remained elusive. In 1985, they paused to survey the literature. Late that year, Bednorz came across a paper by French researchers that described a perovskite of barium, lanthanum, and copper. The French team was interested in its catalytic properties at higher temperatures, but Bednorz realized that it fitted his and Müller's conceptual model perfectly.

Bednorz immediately set about fabricating samples of the Ba-La-Cu oxide. But other duties in the lab and a vacation during January kept him from testing the samples until late January 1986. As he described later in his Nobel lecture, Bednorz cooled down a sample connected to a probe to measure resistivity. At first, the sample appeared to behave like a conductor; then, at 11 K, the resistivity dropped away. Over the next 2 weeks, the pair repeated the experiment over and over, varying the composition of the oxide until they had a material in which the resistivity reliably dropped at 35 K. That was incredibly high by the prevailing standards of superconductivity. But they were well aware that the field was littered with similar claims that could never be reproduced. Nevertheless, given the importance of the discovery, Bednorz and Müller published a paper in the journal *Zeitschrift für Physik* describing "Possible High T_c Superconductivity" before they were 100% sure.

They were missing a crucial piece of evidence: the Meissner effect, the ability of a superconductor to expel all magnetic flux from its interior. The pair had an agonizing wait for the delivery of a DC SQUID magnetometer to perform the magnetic measurements. In September, the machine was installed in the Rüschlikon lab. Bednorz, with the help of Masaaki Takashige, a Japanese researcher visiting Rüschlikon for a year, measured the material's magnetic susceptibility and confirmed the Meissner effect at about the same temperature as the resistivity drop. Bednorz and Müller were now confident that they had found a new class of superconductor and published their new results in *Europhysics Letters*.

Heating up

As the pair expected, Bednorz says, their early talks about the discovery drew a fair amount of skepticism. In late November, however, newspapers reported that Shoji Tanaka's group at the University of Tokyo had successfully repeated their experiments, and Paul Chu's team at the University of Houston in Texas quickly added further confirmation. Chu went on to boost the T_c to 50 K by putting the sample under hydrostatic pressure and then, by replacing the lanthanum with yttrium, achieved superconductivity at the unimaginably high temperature of 93 K.

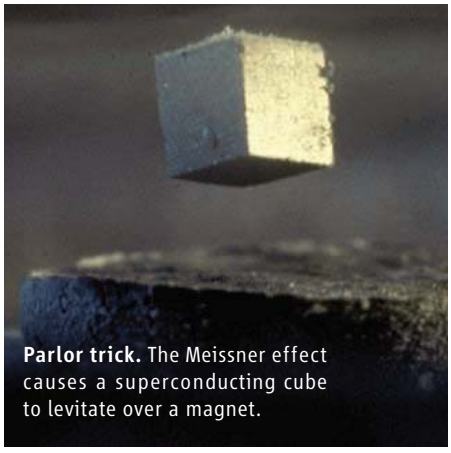
Suddenly, superconductors were the hottest show in town. Dozens of groups were replicating the IBM work and trying different oxides. "It was an exciting time," says Bednorz. Such was the flood of new work that at the March 1987 meeting of the American Physical Society in New York City, a special evening session was hastily organized to hear some of the new work. Organizers allowed researchers just 5 minutes each for their pre-

sentations, as 1800 people crammed into a conference room designed for 1100 and another 2000 watched outside on TV screens. This session, dubbed the "Woodstock of physics," continued into the early hours of the next morning.

While Müller attended the New York meeting as a guest speaker, Bednorz was invited to a meeting of the German Physical Society at his alma mater, the University of Münster. The superconductivity session was so crowded that he had trouble getting in. When he politely asked the person blocking the doorway if he could get through, he was told: "Look, we all want to get in." Bednorz says there followed a period of almost constant travel, mostly to the United States and Japan. He gave 52 talks in 9 months. "It was hard to get any work done," he says.

After receiving the Nobel Prize in physics in December 1987—the shortest gap between discovery and award of any Nobel—things settled down a bit. Bednorz and Müller returned to characterizing the materials and trying other combinations of metals in their perovskites. "We had modest success, but others were quicker," Bednorz says, and the big advances happened elsewhere.

In the early 1990s, Bednorz began experimenting with growing perovskites in thin films on various substrates. They were partly searching for potential applications in electronics, but the technique of growing superconducting oxides by epitaxy—depositing them layer by layer with evaporated atoms—also opened avenues for basic research. Just as Chu's group had boosted T_c with hydrostatic pressure, Bednorz and colleagues studied how T_c changed when they put the superconductor under compressive or tensile stress by using a substrate in which the period of the lattice was slightly smaller or larger than that of the superconducting oxide. Also, by growing a superconductor in a layered semiconductor structure, they could add or remove charge carriers from the oxide to see what effect that had on its conduction.



Parlor trick. The Meissner effect causes a superconducting cube to levitate over a magnet.

In the years following the discovery, Müller collaborated on superconducting applications with researchers at Los Alamos National Laboratory in New Mexico as well as at the firm American Superconductor in Westborough, Massachusetts. He has also remained active in unraveling the mechanism behind their discovery, sticking to his original thesis that a quasi-particle in the crystal lattice called a polaron—an electron and the deformed and polarized lattice around it—is key to the process.

Most superconductivity theorists now believe that such lattice vibrations don't play a role in high-temperature superconductivity. But Müller maintains that the superconducting layer in the perovskites is not homogeneous and that small areas within it harbor polarons, and he cites some experimental results to support his case. "Experimentalists are leading now. Theorists should listen for a bit," Müller says. Paul Grant, an emeritus IBM researcher at the Almaden research center and longtime colleague of Müller, says the idea is worth watching. "He's definitely in the minority, but it's hard to see where he's really going wrong," Grant says. "If he's right, he deserves another trip to Stockholm."

Hot ticket. Grand Ballroom, New York City's Hilton Hotel, 18 March 1987: Physicists cram into the impromptu session, known as the "Woodstock of physics," where dozens of early results are presented.

CREDITS (TOP TO BOTTOM): IBM ZURICH RESEARCH LABORATORY; AIP EMILIO SEGRÉ VISUAL ARCHIVES

Beyond superconductivity

In the mid-1990s, Bednorz changed tack. "I went back to my origins, insulating perovskites, the field that brought myself and Alex Müller together," he says. He is now working on insulating materials that can be momentarily converted into a conducting state with an injection of charge carriers. That's useful because short voltage pulses can flip such materials between two resistance states, each of which is entirely stable on its own and doesn't need any power to keep it that way. If that can be achieved on a minute scale, the material could form the basis of a nonvolatile memory, a chip that remembers data when the power is switched off—a major goal for the computer industry. Bednorz and his colleagues have demonstrated switching in cells as small as 100 by 200 nanometers. "If we can get to these dimensions, with single cells working reliably, it will be very competitive," he says.

Müller, meanwhile, became influential in Swiss science policy circles, and he began to press the government to build a national synchrotron facility. To pursue that goal, a group formed around him that became known as the "Alex Müller Committee," says Leonid Rivkin of the Swiss Light Source, which finally opened its doors in 2001. "I try to help science in such ways," Müller says.

Nine years ago, Müller retired from IBM and moved completely to the University of Zurich, where he had held a part-time position for some years. At age 80, he is still active, writing papers and giving talks. He's got a hunch that sooner or later researchers will come across an entirely new class of high-temperature superconductors. "It's highly unlikely that there isn't another class," he says, adding that he has some ideas but declining to name them.

Grant respects the two researchers highly. "Georg's persistence in pursuing Alex's ideas was key," he says. And Müller's impact on research at IBM has been huge. "He identifies good problems, places to explore, and good people." While Müller headed the physics section at Rüschlikon, he not only won a Nobel Prize but also hired three other researchers who became Nobelists. "All that occurred because of Alex Müller." **–DANIEL CLERY**

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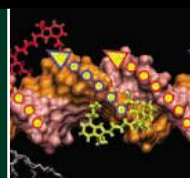
LETTERS | BOOKS | POLICY FORUM | EDUCATION FORUM | PERSPECTIVES

LETTERS

edited by Etta Kavanagh

Why Aren't There More Scientists Advocating for Funding?

THE FUNDING OUTLOOK FOR BIOLOGICAL SCIENCES IN THE UNITED STATES is bleak; success rates are in the single digits for some grant programs at NIH and NSF. Yet, when the Federation of American Societies for Experimental Biology (FASEB) recently sent out a policy alert to its 80,000 members urging support for a congressional measure to increase the NIH budget, only 8000 members responded. Why? The following are a few explanations for this passivity we've heard over the years and why they don't hold up.

Lack of time. Of course, scientists are busy and must do science. However, if every scientist devoted just an hour per month to advocacy, the science funding condition would likely be much improved.

Ignorance. Scientists may not know how to advocate or what to do. Many scientific societies have staff that can help. A little research and a few conversations with them on how to engage in advocacy is all that is needed to get started. Use these resources!

Someone else is doing it. You may think that your personal involvement isn't necessary, since some societies may be engaged. However, there is no substitute for a scientist in a meeting with a legislator. No legislator ignores a constituent. Contacting your legislator should be a regular item on your monthly "To Do" list.

It's too big a job and I'm only one person. The problems of science funding may seem insurmountable. However, the power of a single individual to bring about change has been amply demonstrated throughout history, including by such scientists as Galileo, Charles Darwin, Albert Einstein, and Rachel Carson.

Elitism. Unfortunately, some scientists feel that politics is less ethical than science, and that politicians are not as smart as they are. Thus, getting involved in politics is a step down for them, both morally and intellectually. If getting involved out of a sense of civic duty is not enough motivation, keep in mind that congressmen control our "purse strings" and, thus, it is in our self-interest to do so.

Politicians won't listen to me. Most legislators don't know many scientists and often will value a meeting with you because of your expertise. One of us had a meeting with a prominent senator that went on for well over an hour (such meetings typically last 10 minutes). The senator's comment at the end of the meeting was very telling: "I rarely have the opportunity to talk with a person like you, Dr. Wells." Thus, don't be intimidated. Most legislators will support science if they know that their constituents care.

I can't afford it. Most federal legislators are in Washington part-time and spend the remainder of their time at their home base. Make that local appointment; it will only cost you a bit of time.

I'm not allowed to advocate. Absolutely false. Publicly funded scientists do not relinquish their rights to free speech. Use your own computer at home and your own time. Scientists do not give up their right to free speech because they receive federal funds.

So what's your excuse? Unfortunately, scientists cannot depend on Congress to adequately fund science. We (biologists, physicists, chemists, mathematicians, etc.) must work together. Alternatively, will you wait to act until your lab is unfunded? Science and our country need us.

ROBERT D. WELLS[1] AND PETER FARNHAM[2]*

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Fighting Waterborne Infectious Diseases

IN HIS PERSPECTIVE "WATERBORNE INFECTIOUS diseases—could they be consigned to history?" (Special Issue on Freshwater Resources, 25 Aug., p. 1077), A. Fenwick presents a hopeful outlook, based principally on drug effectiveness and distribution, for gaining control over serious waterborne infectious diseases in the developing world. Although treatment is an effective first step, we issue a cautionary note regarding drug-based strategies as the sole means to eradicate transmission or even to suppress it sustainably to levels below the threshold of concern.

In China, the anti-helminthic drug praziquantel has been the principal tool, together with the molluscicide niclosamide, for notable progress in the control of the *S. japonicum* parasite in humans and domestic animals (*1*–*3*). However, in recent years, the disease has re-emerged in formerly endemic areas where transmission had earlier been terminated (*4*, *5*), leading to widespread recognition that a more comprehensive strategy, including environmental interventions, will be necessary to achieve a long-term solution in China (*6*).

The decrease in disease burden "if exposure to a risk factor were reduced, not to zero, but to some achievable level" by environmental means has recently been estimated to be 100% for schistosomiasis, 66% for lymphatic filariasis, 42% for malaria, and 100% for intestinal nematode infections (*7*). Although

CREDIT: JOE SUTLIFF

these estimates do not diminish the importance of drug therapy, they illustrate that these diseases are susceptible to environmental controls. Moreover, the large increase in drug use required even for adequate control of morbidity for these diseases heightens concern over the potential for drug resistance.

In the same issue, Bill Gates was quoted with reference to the economic dimension of large-scale treatment for HIV/AIDS infections: "Treatment without prevention is simply unsustainable" ("At International AIDS Conference, big names emphasize big gaps," J. Cohen, News of the Week, 25 Aug., p. 1030). We agree. In the case of waterborne infections, prevention often begins with clean water and improved sanitation. It is essential that policy-makers remain committed to these traditional public health measures and not rely solely on the promise of vaccines and inexpensive drugs.

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Response

I THANK SPEAR *ET AL.* FOR HIGHLIGHTING THAT my optimism for gaining control over serious waterborne infectious diseases in the developing world is based principally on drug effectiveness and distribution. Their cautionary note about chemotherapeutically based strategies achieving eradication, or even breaking transmission, reads rather more into my optimism than was there. My optimism is directed toward reduced morbidity rather than eradication of infections.

However much I support chemotherapy for morbidity control, I have stated that eradication cannot be achieved without significant and dramatic improvement in socioeconomic status (*1*). Spear *et al.* state that these diseases are susceptible to environmental controls (*2*). This may be true, but I believe that the degree of environmental controls needed in sub-Saharan Africa is currently unobtainable in many settings. I also believe that the risk of resistance being caused by the large increase in drug use to achieve the control of morbidity is no reason not to use the existing tools to treat those unfortunate people infected with lymphatic filariasis, onchocerciasis, or schistosomiasis. However, monitoring for drug resistance must accompany mass drug administration campaigns (*3*).

Bill Gates's statement that "[t]reatment without prevention is simply unsustainable" may be correct, but for neglected tropical waterborne infections, the cost is so minimal at $0.50 per person per year that it is cost-effective. Exposure to parasitic infections is not in any way to be blamed on individuals so poor that they are dependent on surface water. Thus, although I agree that policy-makers should remain committed to all available pub-

lic health measures, I believe that in the short term, they must rely on the currently available donated and inexpensive drugs.

ALAN FENWICK

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Debating the Worth of NCCAM Research

MARCUS AND GROLLMAN MISS THE MARK IN their critique of the National Center for Complementary and Alternative Medicine (NCCAM) ("Review for NCCAM is overdue," D. M. Marcus and A. P. Grollman, Policy Forum, 21 July, p. 301). We believe that NCCAM, under the leadership of Stephen Straus and Margaret Chesney, has made remarkable progress in laying the groundwork and advancing rigorous research in complementary and alternative medicine (CAM). They have brought definition, a conceptual framework, strategic plans and goals, and scientific standards to the field of CAM research.

The processes through which proposals are submitted, reviewed, funded, and managed are all consistent with standard NIH practice. The disciplinary diversity of the NCCAM study section members and NCCAM councils is in keeping with the breadth of CAM research. The broad representation is also consistent with current practice in other centers and institutes. NIH advisory bodies regularly include members who are grantees, and well-tested procedures are in place for managing conflicts of interest. The same procedures are used for the study sections and advisory councils for NCCAM.

Marcus and Grollman's comment that the NCCAM research agenda is shaped more by politics than by science is gratuitous, as is their suggestion that the Institute of Medicine (IOM) report, *Complementary and Alternative Medicine in the United States* (*1*), was flawed because some of the members of the panel were NCCAM grantees. In fact, like NIH, the IOM has procedures for recognizing and managing conflicts of interest. Those of us who participated in it were very mindful of any potential conflicts of interest and guarded against them in our deliberations. Further, the report was carefully reviewed by external, independent reviewers before publication.

Because CAM is already in the public domain, used by millions of people at a cost of billions of dollars each year and with health effects that largely have not yet been scientifically evaluated, it is appropriate that a significant focus be on clinical research. As is true in clinical trials with new conventional drugs, we should expect that many trials of CAM treat-

Letters to the Editor

Letters (~300 words) discuss material published in *Science* in the previous 6 months or issues of general interest. They can be submitted through the Web (www.submit2science.org) or by regular mail (1200 New York Ave., NW, Washington, DC 20005, USA). Letters are not acknowledged upon receipt, nor are authors generally consulted before publication. Whether published in full or in part, letters are subject to editing for clarity and space.

ments will not show definitive efficacy, and as with most research on understudied agents, multiple studies are often needed to develop a research base adequate for mature judgment concerning efficacy or the lack thereof. We need to be patient and use our best tool, that is, science, to understand and evaluate these widely used health practices. We believe that NCCAM has established a standard not for advocacy, but rather for rigorous objectivity.

SUSAN FOLKMAN,[1] BRIAN BERMAN,[2] STUART BONDURANT,[3] DAVID EISENBERG,[4] AVIAD HARAMATI,[5] MARY JO KREITZER,[6] FREDI KRONENBERG[7]

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Reference

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Response

THE MAIN POINT OF THE LETTER FROM FOLKman *et al.* and the response to our Policy Forum by S. E. Straus and M. A. Chesney ("In defense of NCCAM," Policy Forum, 21 July, p. 303) is that NCCAM uses standard NIH procedures for review of proposals, appointments to advisory and review groups, and management of conflict of interest. That is formally true but misleading. Because of its charter, NCCAM advisory and review groups include many individuals whose scientific credentials would not qualify them for appointment to other NIH institutes. Of greater importance, the continued funding of poor-quality proposals refutes Straus and Chesney's claim that NCCAM applies the same review standards as other NIH institutes.

Except for Bondurant, the signatories of the Folkman *et al.* letter all hold leadership positions in CAM or integrative medicine centers supported by NCCAM. Bondurant is a senior academic officer at Georgetown University Medical Center, which has a CAM center, and he was chairman of the IOM Committee that issued the report on CAM. Berman, Eisenberg, and Folkman also served on the IOM committee.

The NCCAM appropriation for 2005, $123 million, understates NIH expenditures on CAM research. In 2004, the NCCAM budget was $117.8 million, and the total NIH expenditure on CAM research was estimated at $305 million, much of which represented projects that were co-funded by NCCAM and other institutes. NCCAM recently announced the creation of five new centers that will conduct research on multicomponent traditional African and Chinese herbal medicines. Each center will receive approximately $1 million per year, which is the equivalent of 20 RO-1 research grants. Some of this research is meritorious, but much of it is not.

An independent review is urgently needed to bring the evaluation of proposals by NCCAM into line with the rest of NIH and to ensure that the limited funds available for biomedical research support the best science.

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EVOLUTION AND DEVELOPMENT

A Kernel Bears Fruit

Detlev Arendt

How did the different courses of development that we observe today come into existence? What can the development of extant organisms teach us about evolution? In his latest book, *The Regulatory Genome*, Eric Davidson addresses the interrelation between evolution and development, the core matter of the field of evo-devo (*1*). Davidson, a developmental biologist at Caltech, offers the perspective of the world's leading expert on the gene regulatory networks that control animal development. These networks consist of huge sets of regulatory genes that control one another's expression as well as the expression of downstream effector genes via so-called cis-regulatory elements (to which the transcription factors bind). How did evolution shape these gene regulatory networks? And what can we learn about metazoan evolution from the dissection and comparison of these networks? Davidson's answers to these questions represent the main conceptual advance of this excellent book. He categorizes the distinct components (subcircuits) of gene regulatory networks in animal development according to their function and to their different degrees of evolutionary conservation, and he proposes a scenario to explain how these distinct types of network subcircuits have emerged in animal evolution.

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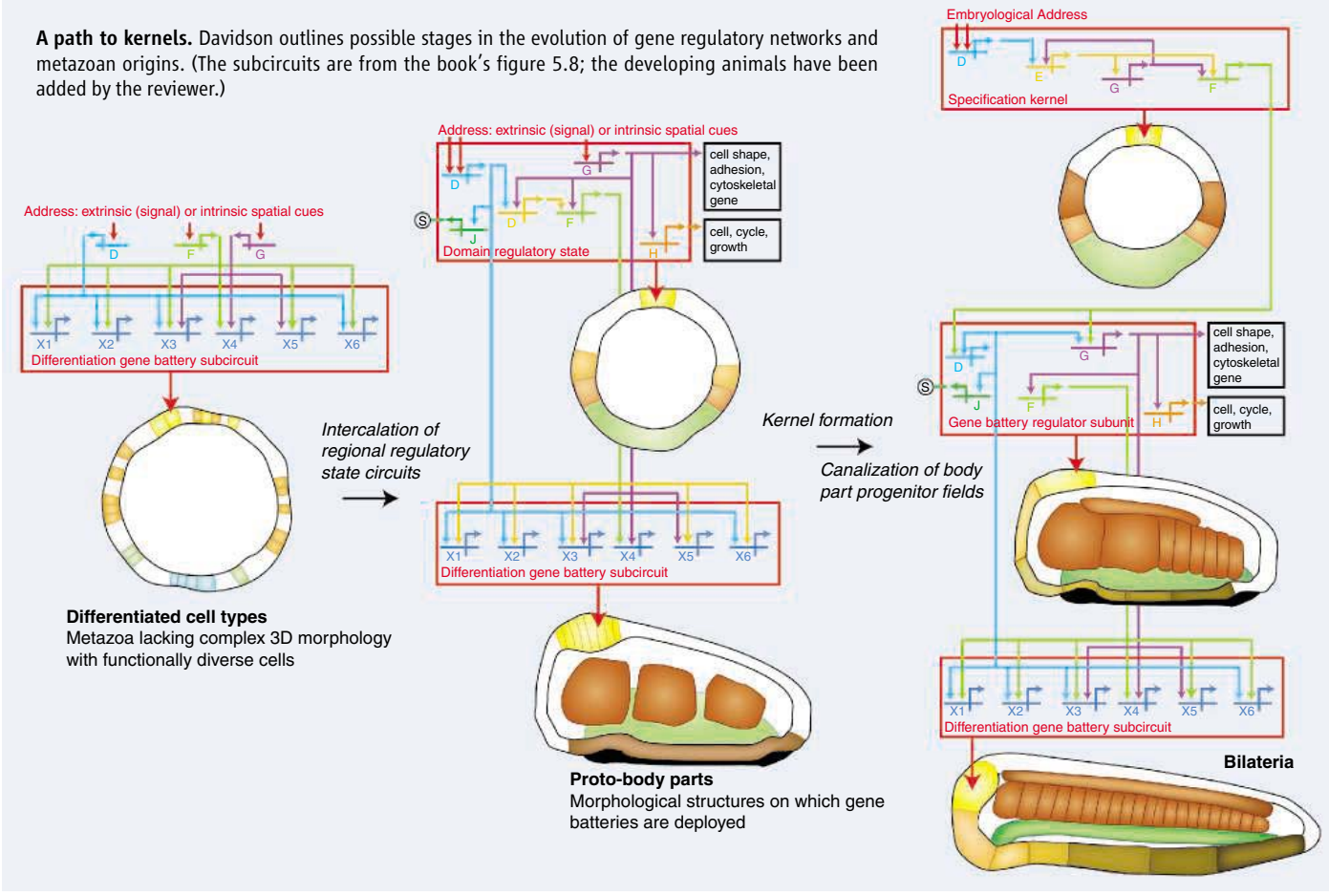
The Regulatory Genome
Gene Regulatory Networks in Development and Evolution

by Eric H. Davidson

Academic Press (Elsevier), Burlington, MA, 2006. 301 pp. $69.95. ISBN 0-12-088563-8.

In the book's first three chapters, Davidson follows up concepts and ideas that he has previously put forward in reviews and a book (*2*–*4*). He begins by providing a detailed account of the architecture and functioning of the cis-regulatory gene regions, which represent the basic nodes of the gene regulatory networks. Davidson then outlines the principles of animal development as a conceptual framework for understanding the activity of gene regulatory networks. His views on animal development have already inspired more than one generation of developmental biologists (including myself). In essence, through a stepwise process, development subdivides the embryo into territories, subterritories, and "progenitor fields" that make up a given body part of the adult animal. This stepwise subdivision is accomplished by gene regulatory network subroutines called subcircuits that consist of small sets of genes and cis-regulatory elements. The interplay between spatial subdivision and regulatory subcircuit activity is schematized in the figure, where different colors label distinct embryonic territo-



A path to kernels. Davidson outlines possible stages in the evolution of gene regulatory networks and metazoan origins. (The subcircuits are from the book's figure 5.8; the developing animals have been added by the reviewer.)

CREDIT: ADAPTED FROM FIG. 5.8 IN *THE REGULATORY GENOME*

ries and progenitor fields. The material covered in these first three chapters is essential for understanding the subsequent two chapters on the organization and evolution of gene regulatory networks.

In "Gene Regulatory Networks for Development," the book's key chapter, Davidson presents a comprehensive survey of the logic, motifs, and parts of gene regulatory networks in development and establishes standards for their structural analysis. He offers a glossary that summarizes general designations and the parts of a developmental gene regulatory network. Judging from various lectures at recent conferences, some of the terms he coined have already reached buzzword status in the evo-devo community. For example, as initially used in informatics, "kernel" refers to the core of an operating system. By analogy, Davidson uses kernel for a highly conserved subcircuit of transcription factors that have a key function in animal development—such as the specification of a progenitor field from which a given body part develops. Acting in unique, kernel-specific combinations, the constitutive transcription factors control one another's expression. In the absence of any one of these factors, the entire network necessarily malfunctions such that the entire body part fails to form. This explains the kernels' "shocking" evolutionary conservation over more than half a billion years of metazoan evolution. As the gold standard for a kernel, Davidson presents a conserved part of the endomesoderm specification network shared by starfish (*Asterina miniata*) and sea urchin (*Strongylocentrotus purpuratus*). In addition to the highly conserved kernel subcircuits, Davidson distinguishes among the more variable plug-ins (such as signaling cassettes), the input and output switches (such as the *Hox* genes) that decide whether and how another subcircuit is activated, and the differentiation gene batteries that establish the final cell types. After discussing such general structural properties, the author considers these four different forms of subcircuits as they are exemplified in a set of developmental networks that have been extensively studied. First and foremost among these is the sea urchin endomesoderm network, which has been a focus of the author's own research. His other examples are mesoderm specification in the *Xenopus* embryo, dorsal-ventral specification in the *Drosophila* embryo, specification in pancreatic β-cells, and terminal differentiation in *Caenorhabditis elegans* taste neurons.

The final chapter, on the evolution of gene regulatory networks, is the most speculative and most stimulating. Here, Davidson outlines a possible evolutionary origin of kernels, which I summarize in the figure. The evolutionarily oldest components of gene regulatory networks would be the differentiation gene batteries that defined and shaped the limited number of cell types already present in early metazoans, when complex morphologies did not yet exist (left panels in figure). In these early animals' development, the differentiation gene batteries directly responded to simple spatial cues via rather simple gene regulatory network subcircuits. The second stage in metazoan evolution was the assembly of an increasing number of cell types into body parts such as gut or brain (middle panels). In the developing animal, this step brought about the subdivision into territories, which were specified via newly evolving regulatory subcircuits that were intercalated into the preexisting network. Typically, these subcircuits recruited transcription factors already present as part of the differentiation gene batteries. In a third stage (right panels), the animal body parts and, concomitantly, the territorial subdivision of the developing embryo became more and more elaborate until finally the underlying subcircuits were locked down into kernels that could no longer be changed without deleterious consequences. According to Davidson, this "triumph of the bilaterian versions of animal body plans" was in place sometime before the Cambrian and has persisted, constraining metazoan evolution ever since with tremendous success.

The proposed link between the evolution of kernels and the evolution of bilaterian body plans is exciting, but it awaits validation through more comparative analyses of gene networks. As Davidson himself concedes, "for the identification of kernels … an overwhelming feature of the evidence thus far is its thinness." For example, he identifies as a putative pan-bilaterian kernel the heart specification subcircuits in *Drosophila* and the mouse. Although some core transcription factors are conserved between the two, major interactions are carried out in "slightly different ways" involving different transcription factors, which indicates some degree of plasticity in kernel composition. Davidson also discusses as possible kernels the sets of transcription factors that play conserved roles in nervous system regionalization (*5*–*7*). For some of these sets, conserved mutually repressive interactions have indeed been documented (*8*, *9*). But comparative network data are still scarce for most nervous system regionalization genes that are conserved across bilateria or deuterostomes. It is fair to say that we have good evidence for a collection of conserved network "drivers," which in Davidson's terminology are transcription factors that show spatially restricted expression and convey regional identity. Whether or not these indeed assemble into kernels will have to be determined by future research.

The Regulatory Genome offers evo-devo aficionados an intellectual masterpiece to praise or to pan—but impossible to ignore. Although there is clearly still much to learn about the evolution of gene networks and how these in turn constrain evolution, Davidson has placed a cornerstone for the comparative analysis of gene regulatory networks. Further research in this rather fresh field promises to help delineate the links between development and evolution.

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10.1126/science.1134545

BIOGRAPHY

A Fresh Look at Darwin

Janet Browne

On an autumn day in Chicago in 1959, exactly 100 years after the publication of *On the Origin of Species*, a large and appreciative audience of biologists attended an evolutionary musical called *Time Will Tell*. Earlier in the day they had heard Julian Huxley, one of the architects of the new evolutionary synthesis, declare that religious belief was merely an adaptive social feature of early mankind. That same year a partial reenactment of the *Beagle* voyage took place and plans were announced for a Darwin memorial park on the Galápagos Islands that meshed with international pressure on Ecuador to restrict commercial fisheries around the archipelago.

Anniversaries, commemorations and the public theatre of ceremonies, lectures and

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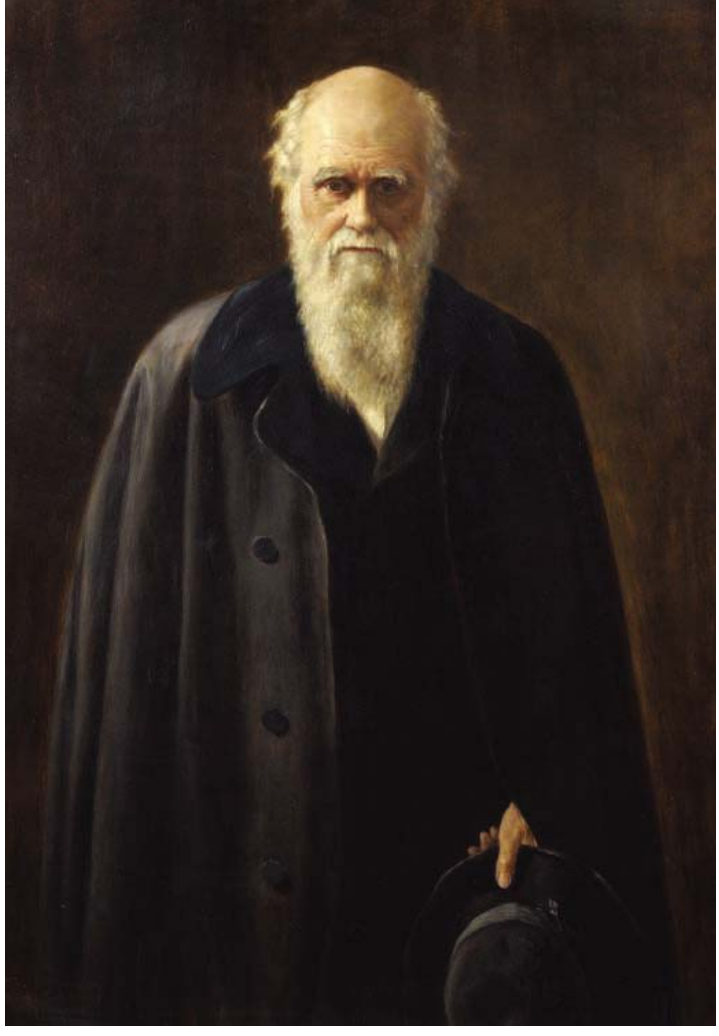

prize-givings are, of course, big business and have long been acknowledged as strategic events for promoting culturally and scientifically significant agendas. The fact that Charles Darwin has been as important after his death as during his lifetime comes as no surprise. The theory of evolution by natural selection, jointly proposed by Darwin and Alfred Russel Wallace, is rightly regarded as the foundation stone of modern biology and underpins much of how the human race has come to comprehend itself. As commonly understood, these writings challenged everything that had previously been thought about living beings, firing hot debate in the intellectual, social, and religious transformations of the 19th century. Yet in 1959 religious controversy over Darwinism was relatively muted. The achievements of 20th-century biology were obvious for all to see. Intellectuals like Julian Huxley used the occasion to praise secular humanism and the rigor, honesty, and dedication shown by practitioners of the new laboratory sciences. Biologists demonstrated that it was entirely possible for a scientist to be a nonbeliever and a valuable member of society. Noted clerics in Europe and North America preached reconciliatory sermons. Nowadays, with 2009—a time that will be used to celebrate the bicentennial of Darwin's birth and the 150th anniversary of publication of the *Origin of Species*—glimmering on the horizon, it is clear that the shape of the debate has materially changed. Would an evolutionary musical be so loudly applauded today given the other issues currently at stake?

The Reluctant Mr. Darwin
An Intimate Portrait of Charles Darwin and the Making of His Theory of Evolution
by David Quammen
Norton, New York, 2006. 304 pp. $22.95, C$30. ISBN 0-393-05981-2.

While the political and cultural controversy surrounding evolution has flared again, there has been a steady stream of accounts of Darwin's life and ideas written for a broad audience. The latest such work, David Quammen's *Reluctant Mr. Darwin*, is a complete delight. Renowned as an author and traveler, Quammen earned well-deserved acclaim with *The Song of the Dodo*, in which he wove together accounts of island biogeography, species extinctions, and Wallace's travels in the Malay archipelago (*1*). He brings the same flair and fluency to this captivating biographical essay. The book is fresh and original, even to those who have explored other biographies of Darwin published over the last decade or so; readers will find it to be just as insightful as many a heavier tome.

Quammen's aim is to open up Darwin's character as a thinking man. He does not take a conventional chronological view from birth to death, nor is he particularly engaged with documenting the emergence of evolutionary biology as a dominant mode of thought. The book is more of a personal reflection on those aspects of Darwin's story that have intrigued him, perhaps as he moved through remote places documenting nature's fecundity or shadowing field workers to describe their adventures and ideas. Quammen leads us through the main features of Darwin's life and thought after his return from the *Beagle* voyage, building up to *Origin of Species* (1859). These events are framed by a couple of fascinating chapters, front and back, that set Darwin's achievements in modern context and reveal some of the reasons for the powerful respect that practicing field naturalists and biologists feel for him today.

There are many insights along the way. Darwin's time-consuming work on barnacles is described with a deep understanding of why taxonomy matters. Darwin's interactions with Wallace are given clear-eyed examination: the subtle combination of panic, generosity, admiration, and regret that each man displayed toward the other is brought newly alive. And the skills of a novelist creep in. Quammen evokes a pleasing image of Darwin playing billiards—a known historical fact that in Quammen's hands suddenly turns the ill and tormented figure who was slaving away writing *Origin of Species* into a real person, smoking cigarettes, laying down his cue, joking about his "abominable volume," and telling his friends how refreshing it is to idle the day away.

For historical scholars, this has always been one of the most difficult paradoxes: how to connect the man remembered by his friends and family as a warm, even jolly, figure with the nervous invalid documented in contemporary records and the incisive author of *Origin of Species*. Quammen's gift is to describe these aspects of Darwin's character without resorting to Jekyll and Hyde imagery. Not only does Quammen enrich our understanding of what it must have been like to be Darwin (a quiet, humane, and determined thinker running deliberately against the Victorian intellectual and cultural stream), but he also shows the lasting brilliance of the theories put forward nearly 150 years ago and explains the great affection with which Darwin is still regarded by naturalists today. If you are going to buy only one book to commemorate Darwin in 2009, *The Reluctant Mr. Darwin* could surely be it.

Reference

1. D. Quammen, *The Song of the Dodo: Island Biography in an Age of Extinctions* (Scribner, New York, 1996).

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RESEARCH FUNDING

NIH in the Post-Doubling Era: Realities and Strategies

Immediate- to long-term approaches are discussed that will minimize the negative impacts of current budget constraints and still preserve the NIH mission.

Elias A. Zerhouni*

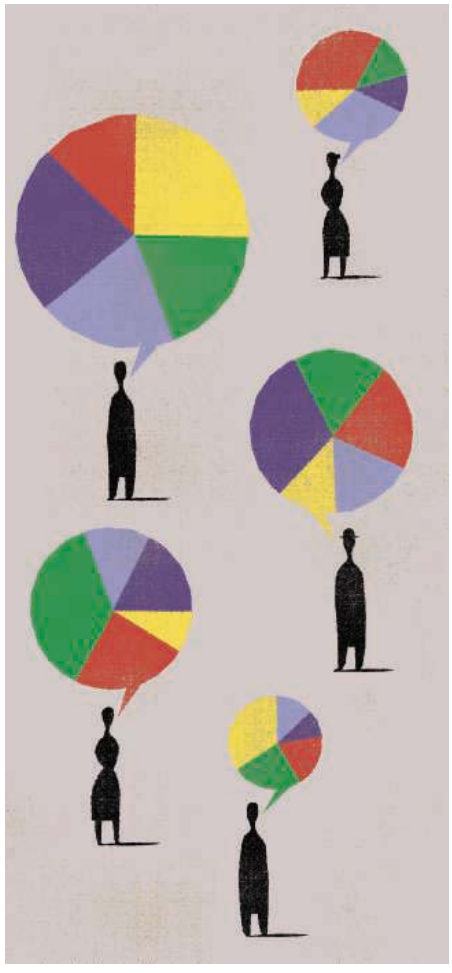
This has been a challenging year for the National Institutes of Health (NIH) and the biomedical research community. An extraordinarily tight federal budget is eroding the growth of NIH at a time when opportunities for scientific progress and advances in human health have never been greater. As I talk to scientists and administrators throughout the country, the anxiety is palpable. I share these concerns. I am most deeply troubled about the impact of this difficult situation on junior scientists, and on the ability of established investigators to maintain their laboratories.

Enhanced online at www.sciencemag.org/cgi/content/full/314/5802/1088

To engage in a productive dialogue about the future, I provide here some data, share my perspective about the main causes of the present situation, and outline the actions we are taking to reduce the very real strain on our scientists.

Realities

Many scientists are dismayed that it is more difficult to get funded today than it was before the NIH budget doubled. What can explain this apparent paradox? The core reason is the increase in the number of new applications and applicants for NIH grants (see figure, p. 1089). In 1998, NIH received 24,151 applications for new and competing research project grants (RPGs) (*1*); NIH expects to receive over 46,000 in 2006 and over 49,000 in 2007. The doubling in the demand for grants is primarily due to a large increase in the number of new scientists applying for grants. In 1998, there were about 19,000 scientists applying for competing awards. In 2006, NIH expects to receive applications from approximately 34,000 scientists and forecasts that over 36,000 scientists will apply in 2007. Remarkably, the largest surge in demand for grants occurred at the end of the doubling period and continues today. This "perfect storm"—the imbalance between supply and demand for grants—is the fundamental reason for the painful circumstances in which we find ourselves.



The principal cause of this remarkable growth in grant demand is the unprecedented expansion of research capacity across the country that began in 1999. Stimulated by successive administrations' and Congress's calling for more research on emerging health issues, academic institutions responded. Using philanthropic, local, and state resources, as well as loans, they expanded the scientific infrastructure and workforce to address the growing scope and complexity of our scientific challenges. For example, the American Association of Medical Colleges projects that an estimated $15 billion have been committed to new research facilities between 1998 and 2007, compared with $3.2 billion between 1990 and 1997. Allowing for the lag time necessary to build facilities and train scientists, this expansion is now being felt in the form of a rapid surge in applications. It should not go without mentioning, however, that this increased investment by our research institutions is resulting in the development of entirely new fields of research, leading to an acceleration of the pace of promising research advances across the entire spectrum of the biomedical and behavioral sciences. This is just what the nation wants and needs.

Unfortunately, our ability to sustain this expanded research enterprise is now at risk. Some of the tension is due to inflation. Since 1998, the average size of RPGs grew by about 40%, and NIH budgets have not kept pace with biomedical research and development inflation since 2003 (*2*).

Increased demand, inflation effects, and flat budgets are the main drivers of today's challenges. It has been suggested that decreased success rates are the result of NIH's excessively shifting its emphasis to applied research and clinical trials through large solicitations and projects at the expense of unsolicited, investigator-initiated basic research. This is simply not the case. In 1998, 54% of the total budget of NIH was dedicated to basic research, 40% to applied research (including clinical trials), and 6% to infrastructure programs. Funding for basic science is currently above 55% and is slated to grow beyond 56% in 2007; at which point, applied research will reach 41%, and 3% will be devoted to infrastructure needs.

NIH embraces the importance of investigator-initiated research. We are firmly committed to independent grant mechanisms such as the unsolicited R01. Although the absolute number of requests for applications (RFAs) (*3*) grew during the early part of the doubling, their proportion relative to the NIH budget decreased since the doubling ended. Today, 91% of funded RPGs are unsolicited, compared with 88% in 2003, and 92% in 1998. Of our extramural grants budget, 78% remains dedicated to RPGs (*4*) as compared with 81% in 1998. The difference is primarily due to growth in centers and contract mechanisms to address emerging public health priorities.

It has also been suggested that the NIH

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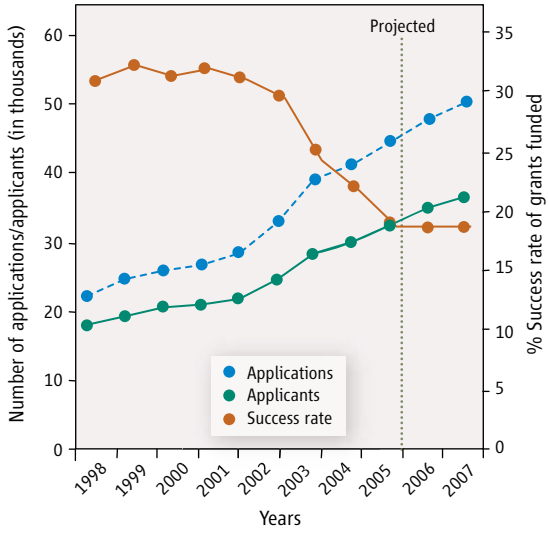
Roadmap (*5*) is a major cause of reduced success rates. In fact, the Roadmap represents only 1.2% of the fiscal year (FY) 2006 NIH budget. The science within the Roadmap is peer-reviewed and very competitive. It is not a monolithic program, but rather supports over 345 principal investigators at 133 extramural institutions through a variety of mechanisms, including R01s. It does not significantly affect overall success rates. The planned Roadmap budget represents a balanced portfolio, with 40% going to basic research; 40% to clinical and translational research; and 20% to interdisciplinary and high-risk research, such as the Pioneer awards.

I believe that any organization of the size and complexity of NIH needs to have an explicit and dynamic process for supporting critical scientific programs that cut across scientific areas and that none of the individual institutes could support on its own. In an era of rapid convergence of (and emerging opportunities in) science, the Roadmap process allows NIH to support innovative and high-risk research, incubate new ideas, and stimulate the development of transforming strategies that can benefit the entire scientific community. To ensure vitality, no initiative will be funded for more than 10 years, with most lasting 5 years. In my opinion, the greatest risk for science is to stop taking risks. The Roadmap process allows NIH to remain responsive even in constrained times. It has been and will continue to be developed through wide consultations with members of the scientific community. The Roadmap process was well received by Congress and the administration and served as an important part of the rationale for NIH's small budget increases in 2004 and 2005.



> "Pragmatic and prudent **steps need to be taken** to minimize the long-term negative impact of the hopefully short-term budget woes."

Strategies

Given these facts, what strategies for the future should we, as a community, consider? Pragmatic and prudent steps need to be taken to minimize the long-term negative impact of the hopefully short-term budget woes. We must develop unified, informed, and proactive strategies.

We need to remain focused on our core values and to pursue our fundamental mission of discovery—translating new knowledge into tangible benefits for the American people. This must remain our top priority. This means maintaining, to the greatest extent possible, the ability of scientists at all stages of their careers to continue their work.

Preserving future generations of scientists. Like farmers during difficult times, we should not "eat our seed corn," but protect it. To accomplish this, we have implemented three specific strategies to encourage and support junior scientists and new investigators: (i) Every institute and center is working to ensure that the success rates of new investigators are not disproportionately affected by flat budgets, through various mechanisms such as differential pay line considerations. (ii) Because new investigators may not have the resources to sustain long peer-review cycles, they now receive their critiques within 1 week of review and can apply up to 3 weeks beyond the next receipt date. This dramatically decreases the length of the application review cycle without compromising the rigor of peer review. (iii) Outstanding new investigators still in postdoctoral training may now apply to the Pathway to Independence awards program launched in 2006 (*6*). It will fund 150 to 200 postdoctoral candidates for each of the next 5 years. These scientists will receive up to 2 years of mentored training support, followed by 3 years of R01-level funding, contingent upon securing a tenure-track position with appropriate institutional support and resources. This strategy is designed to retain promising scientists and to give them the opportunity for independent research at an earlier stage of their career. Some institutes and centers are looking to expand this and other pilot programs in the future.

I remain concerned about how long it now takes for a scientist to launch his or her independent research program. Today, the average age at which a scientist receives a tenure-track faculty appointment has increased to 38, and the average age for receiving a first independent award from NIH is above 40. This trend must be reversed, and the new Pathways program is one component of our strategy to do so.

We all agree on the urgency of developing new and better ways of maintaining the attractiveness, joy, and excitement of a research career while eliminating the daunting obstacles and rigid traditions that junior researchers are facing in our academic systems. Now is not the time to discourage young scientists, but to find bold ways of improving their career prospects and opportunities.

Balancing supply and demand. NIH spends more on funding grants today than ever before, but over 80% of its budget is committed to ongoing projects. In any given year, the only resources available for new grants are those that come from ending projects that started 4 to 5 years before, plus any new increase in the overall budget. Our large commitment base, the unexpected budgetary impact of hurricane Katrina, the fact that in 2006 the only funds available were those freed up from grants started when the NIH budget had not reached its peak, along with the growing number of applications, compounded our difficulties. To alleviate the strain, we made the hard decision to reduce the committed budget of existing awards by 2.35% for 2006, freeing up some resources for new and competing awards. In the coming years, as we recycle funds from the higher funding years of 2003 to 2005, more uncommitted funds will become available, and thus more new and competing RPGs can be awarded.

Although strategic priority setting and careful management of these recycled funds will help stabilize success rates, they will not be sufficient to satisfy the much larger demand for grant funding, if the budget remains flat and inflation continues to erode our purchasing power. While these potential solutions will have an impact on NIH as a whole, we recognize that neither the problems nor the solutions fit every institute and

center. This is why we asked each institute and center to adjust its overall portfolio to preserve individual success rates to the extent possible. The difficult decisions that these adjustments require are made in consultation with institutes' and centers' outside scientific advisory councils.

At the NIH level, we are also redirecting priorities. The intramural program, NIH administrative costs, and infrastructure expenses are being kept well below inflation.

> "**... the greatest risk for science** is to stop taking risks."

Given the current environment, Roadmap budgets are also reduced, and no new initiative within the Roadmap can be undertaken unless it fits within the budget agreed to by all the institute and center directors. This means that for the foreseeable future, new Roadmap initiatives can only begin as other Roadmap initiatives conclude.

Improving peer review. Exacerbating current frustrations for investigators and reviewers is the burden associated with the submission of even more grant applications by applicants seeking to improve their chances of success. In 2006, success rates per application fell to about 20%, while the funding rate for applicants was higher at about 25% (*7*). This is due, in part, to the fact that we now receive on average 1.4 applications per applicant, compared with 1.2 before 2003. NIH is reevaluating its review system to reduce the length of the review cycle for all applicants, shorten grant applications, reduce unnecessarily burdensome procedures, and further improve the quality of our peer-review system.

Communicating the benefits of medical research. NIH and the scientific community need to better educate the public about the extraordinary return on investment in the NIH. The value of NIH is so self-evident to our community that we often do not realize that it is not evident to many others. In a survey last year, 73% of Americans could not name NIH as the government agency that funds most of the medical research paid for by U.S. taxpayers (*8*). I have placed a high priority on enhancing NIH-wide public education efforts, and want to commend the efforts that have been made by all stakeholders, but this is not enough. Congress continually asks me to demonstrate the benefits of the NIH doubling to the American people. We testified to this effect numerous times (*9*). For example, the estimated total cumulative investment in cardiovascular research at NIH per American over the past 30 years is about $110, or approximately $4 for each American per year over the entire period. In return, we enjoyed a 63% decrease in mortality due to heart disease. The value to Americans of this increase in life expectancy has been estimated at about $1.5 trillion per year over the 1970–90 period (*10*). This is an impressive return on investment by any measure, even if only a fraction of the gain came from medical research.

Nonetheless, we all need to do better in demonstrating our value to society. Since 83% of the NIH's budget goes to more than 3000 research institutions across the country, it is a shared responsibility to communicate clearly and consistently to the public the linkage between NIH and advances at the local and regional levels.

This renewed effort on communicating the value of NIH to the American public by all stakeholders is critically important. During a recent debate in the House of Representatives about a bill to reauthorize the NIH, the chief sponsor of the bill, Congressman Joe Barton, measured NIH's success in simple terms: "It helps my family. It helps every American family" (*11*). Representatives made uniformly positive comments about the importance of supporting NIH and increasing its budget by 5% per year for the next 3 years. The bill passed by a strong bipartisan vote of 414 to 2 (*12*), a hopeful sign.

Defining a compelling vision for the future. Continued support for NIH will not be based on past performance, but on a shared and compelling vision for a future that serves the fundamental needs of our society. Today, health-care costs are rising at an unsustainable rate. Scientists need to be an intrinsic part of the solution to this problem. Marginal reform of how health care is delivered will not suffice. We need to radically change what is being delivered. There is an urgent need to transform health and medicine from the curative and onerous paradigm of today to the vision of a more predictive, personalized, and preemptive world of health care. The only hope to do so is to further our fundamental understanding of biology and behavior through sustained scientific discovery.

Since 1945, United States success in scientific research and development has been the result of the implicit partnership that exists among academia, the federal government, and industry. In this model, research institutions take the risk of building and developing our national scientific capacity; the federal government, through a competitive peer-review process, funds the best science; and industry plays the critical role of bringing new, safe, and effective products to the public. This strategy is the keystone to sustaining American competitiveness, and must be preserved.

As a community, we are accomplishing a great deal, but we are in particularly difficult times. Although these are very challenging and painful days, I am confident that we will weather this storm. Now more than ever, an informed, proactive, and unified strategy will be key to advancing the science needed to improve the health of the world. I welcome comments and suggestions on how we can come together as a community to achieve this goal.

References and Notes

1. The major funding instruments used by NIH to fund extramural research are financial assistance award grants and cooperative agreement grants. Research project grants are awarded to institutions on behalf of a principal investigator to support medical research activities in the areas that represent both the specific interests and competence of the principal investigators and also the NIH institutes' identified program needs. These are generally initiated by the investigator.
2. Price indices, NIH Office of Budget, http://officeofbudget.od.nih.gov/UI/GDP_FromGenBudget.htm.
3. A request for application (RFA) is the official statement that invites grant or cooperative agreement applications to accomplish a specific program purpose. RFAs indicate the amount of funds set aside for the competition and generally identify a single application receipt date. Applications for R01s and other types of grants submitted in response to RFAs are also known as solicited applications.
4. The extramural grants budget includes research project grants, research centers (grants to support long-term, multidisciplinary programs of medical research), other research grants, such as grants for research career development, and other small grant programs. The extramural grant budget represents 70% of the total NIH budget.
5. NIH Roadmap, http://nihroadmap.nih.gov/.
6. NIH new investigator programs, http://grants1.nih.gov/grants/new_investigators/index.htm.
7. Success rates indicate the percentage of reviewed research project grant (RPG) applications that receive funding. This is computed on a fiscal-year basis. Applications that have one or more amendments in the same fiscal year are only counted once. Success rates should not be confused with funding rates. Funding rates indicate the percentage of competitively reviewed applicants for RPGs that receive funding in any fiscal year. This is calculated by dividing the number of applicants that received an RPG award by the total number that competed for any RPG in the same fiscal year (some individuals apply for more than one RPG).
8. M. Woolley, S. M. Propst, *JAMA* **294**, 1380 (2005).
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11. Statement of Representative Joe Barton (R–TX) on House floor, 28 September 2006.
12. Vote on NIH reauthorization on House floor, 28 September 2006.

10.1126/science.1136931

PSYCHOLOGY

Money Is Material

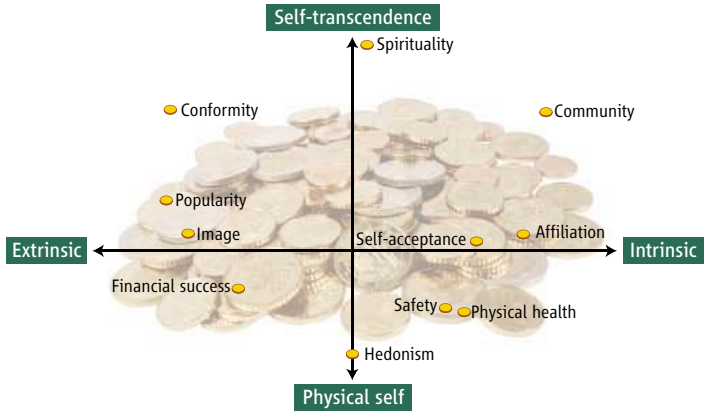
The psychology of money is now being studied experimentally. Even thinking about money changes behavior in reliable ways.

Carole B. Burgoyne and Stephen E. G. Lea

Despite the importance of money in everyday modern life, the psychology of money has until recently received relatively little attention within science, let alone *Science*. But on page 1154 of the current issue, Vohs *et al*. report that even quite trivial exposure to the idea of money (for example, unscrambling phrases about money or reading an essay about money aloud) changes the goals and behavior of test subjects (*1*). These changes occur in the direction of what Vohs *et al*. call "self-sufficiency": Subjects exposed to the idea of money subsequently show a more self-reliant but also more self-centered approach to problem-solving than subjects exposed to neutral concepts. These findings of Vohs *et al.* echo findings in other areas, for example, those of Bargh and colleagues who found that just reading a few words relating to achievement instead of neutral words caused people to outperform controls on a cognitive task (solving word-search puzzles) [see experiment 1 in (*2*)].

Those few researchers who have studied this topic have mostly drawn on the methodological and conceptual tools of sociology and anthropology [see, for example, Belk and Wallendorf (*3*)], rather than those of experimental psychology or neuroscience. This is in part because, on an evolutionary time scale, money is a recent phenomenon, with a history going back no more than a few thousand years, and the forms it takes across history and cultures vary widely (*4*). It seems unlikely that any brain mechanism could have evolved in this time specifically to handle money, so there has been a tendency to treat money as a purely cultural phenomenon for which no scientific account can be given. However, to a biological psychologist, any kind of behavior must be mediated by material (that is, physical) processes, presumably neurochemical events in the brain. Behavior toward money, and the pleasure that people derive from obtaining money, must therefore be mediated in one of two ways. Either it must involve brain processes that evolved to deal with the things money is exchanged for [Lea and Webley (*5*) call this a "tool" explanation of money effects], or it must involve brain processes that money somehow captures in a way that could be maladaptive (they call this a "drug" explanation).

It is all very well to claim in principle that behavior toward money must be mediated by material processes and therefore open to scientific investigation. But what evidence can be produced?



Polar opposites. Two-dimensional representation of the relationships between 11 goal domains. Data are derived from a questionnaire about the importance of 57 different goals given to a sample of 1854 undergraduates from 15 different countries. Note the diametrically opposite placement of financial success and community. [Reprinted from Grouzet *et al.* (*21*) with permission from the authors and publisher (the American Psychological Association).]

First, emerging fields of study such as economic psychology (*6*), behavioral economics (*7*), and experimental economics (*8*) have driven back the orthodoxy that economics could best be studied by purely mathematical and theoretical methods (*9*). Empirical results from these fields have shown that behavior toward money is consistent and predictable, although not always what common sense or economic theory would predict (*10*). The work of Vohs *et al*. stands in this tradition.

Other examples of predictable responses include money illusion, money conservatism, and money taboos. Money illusion refers to the way human decisions are frequently affected by the nominal rather than the real value of money—e.g., they fail to take inflation into account (*11, 12*) and adapt slowly to the values of a new currency unit such as the euro (*13*). Money conservatism is the frequent tendency for people to be disproportionately hostile to currency reforms, even when they are economically desirable: An example is the resistance to the reintroduction of a dollar coin in the United States (*14*). Money taboos are social rules that prevent money from functioning, as it ideally should according to economic theory, as a universal medium of exchange. There are many exchanges where it is not socially acceptable to use money directly (although it is often used in an indirect, disguised way). People in western cultures find it offensive to consider that they may be exchanging sex for money (*15*) or to set a financial value on their children (*16*). Most will not contemplate giving their parents or grandparents money as a gift, although this is perfectly acceptable for gifts traveling down a status hierarchy (*17*).

The second line of evidence that behavior toward money is open to scientific study comes from the rise of neuroeconomics (*18*). This approach is yielding a healthy body of interesting experimental results showing, for example, that separate neural systems are activated when people are offered immediate and delayed monetary rewards (*19*) or fair and unfair offers in a standard economic game (*20*). These results and the example of Vohs *et al*. show that the study of money can be acceptably scientific: Money is, indeed, material.

Our title, however, is a multiple pun. We want to emphasize that the scientific study of money is not just possible but important. It matters for two reasons. First, money is a very large fact in the lives of everyone who lives in a modern economy. Second, the way we respond to that fact makes a difference in our lives. Vohs *et al.* show that merely thinking about money can push people into a narrowly individualistic frame of mind. This provides further experimental support for a position social scientists have been taking for some time now. As an example, the figure shows that across 15 different cultures, "financial success" as a goal is in direct opposition to goals concerning "community" (although less so for poorer cultures) (*21*). Monetizing a

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transaction can change the nature of a social contract: Fining parents who arrived late to collect their children from a day-care facility led (paradoxically) to more parents thereafter consistently turning up late, seemingly content to pay the price for their behavior (*22*).

Being overly preoccupied with money, especially for the "wrong" reasons (*23*), is characteristic of those who score highly on a measure of materialism, and such people tend to be less happy than others (*24*). Given the centrality of money in modern societies, gaining a more comprehensive understanding of the causes and effects of behavior toward money is clearly not just a scientific project; it also has a contribution to make toward understanding, and perhaps enhancing, human happiness and well-being.

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GEOLOGY

Why Do Freezing Rocks Break?

Contrary to common perception, the breaking of rocks is usually not caused by the expansion of water upon freezing.

Bernard Hallet

If you leave a bottle of wine in your freezer a bit too long, you will find it chilled but fractured. This common experience is a result of the volumetric expansion by 9% during the water-to-ice transition, which can generate tremendous pressure in a confined space. One may thus expect freezing water to also fracture rocks. Yet, a different process is likely to prevail in rocks and may also underlie a range of other phenomena. On page 1127 of this issue, Murton *et al.* (*1*) report an integrated experimental and theoretical study that examines rock fracture due to this process under realistic conditions.

The power of the 9% water-to-ice expansion in confined spaces is undeniable, but it may rarely be significant for rocks under natural conditions, because it requires a tight orchestration of unusual conditions. Unless the rocks are essentially saturated with water (*2*) and frozen from all sides, the expansion can simply be accommodated by the flow of water into empty pores, or out of the rock through its unfrozen side. The widespread notion that incipient cracks at the surface of rocks can be wedged open by freezing (as commonly cartooned in textbooks) may also be rarely important in nature, because water can leak out of the cracks, and the ice capping the cracks can push out (*3*).

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The more likely rock fracture process involves freezing but is independent of the water-to-ice expansion. Experiments have shown (*4*) that even liquids that contract upon freezing—most recently argon and helium (*5*)—can cause the expansion of soils and other porous materials. The expansion of moist soils upon freezing results from the growth of ice lenses (known as segregation ice) sustained by a supply of water driven thermodynamically along unfrozen films toward growing ice lenses (*4*). Intermolecular forces that act between the mineral surfaces, ice, and water sustain these unfrozen films and generate pressure between mineral and ice surfaces (*5*). As sub-zero temperatures decrease, the films thin rapidly, thereby restricting water supply to ice lenses, but the maximum attainable pressure increases.

Murton *et al*. (*1*) examine the fracture of limestone samples due to the growth of segregation ice. This process has long been recognized as a weathering mechanism (*4*, *6*), and similar ice growth has been observed experimentally in other rocks (*7*). However, before the study by Murton *et al*., the process had not been subjected to an integrated study involving laboratory experiments and theoretical analyses under realistic temperature and moisture conditions.

How to fracture rocks. Under ideal conditions—ample moisture and mild freezing—intact frost-sensitive cobbles are reduced to fans of rock slivers within decades in Icy Bay, Alaska. Murton *et al.* provide insights into the mechanism by which rocks fracture.

The results reported by Murton *et al.* are in accord with earlier theoretical predictions (*6*) and experimental findings (*8*), which showed that freezing sandstone does not fracture at the nominal freezing temperature of water of ~0°C (as would be expected if it resulted from the expansion of water turning to ice). Rather, it fractures primarily at lower temperatures, which are necessary for substantial pressure within microcracks to develop as segregation ice grows, but not so cold that the unfrozen water films thin so much as to effectively cut off water flow to the growing ice. For this and similar types of rock (see the figure), the temperature range critical for rapid, segregation ice–induced fracture is –3 to –6°C.

This current understanding casts doubt on the long-standing assumption that the 9% volumetric expansion is critical for freezing to fracture rock, and the closely associated notion that the frequency and intensity of freeze-thaw cycles are the main environmental determinants of frost weathering. These cycles may nevertheless be important—not because they cross 0°C, but rather because large temperature gradients arise as the rock is brought into the critical subzero temperature range favorable for water migration and segregation-ice growth. These ideas pave the way to a more fundamental understanding of the effects of rock type and climate on frost weathering (*8*), and more generally, of the basic processes underlying many soils and landforms that are characteristic of cold regions, not only on Earth but also on Mars and other cold planets.

The realization that frost damage in porous materials is, in general, fundamentally related to water being driven thermodynamically into small cracks, where it forms segregation ice, provides a fresh perspective on other forms of rock breakdown. They include those due to moisture variations and salt crystallization at above-freezing temperatures. As Taber (*4*) realized long ago, ice growth in soils is closely analogous to mineral crystallization in rocks. Modeling of frost weathering (*1*, *6*) thus provides strong guidance for studying rock expansion and weathering due to wetting or the growth of salt crystals.

Fundamental insights into liquid water and freezing in confined spaces have recently emerged from studies of the premelting phenomenon (that is, the occurrence in most materials of liquid films on surfaces and interfaces at temperatures far below their bulk melting temperature). In a recent review, Dash *et al.* (*5*) discuss the physics of ice premelting and explore the diverse geophysical manifestations of the basic phenomenon on land, in the oceans, and throughout the atmosphere and biosphere. They discuss briefly the growth of segregation ice in rocks and the resulting fracture that were examined quantitatively by Murton *et al.*

Insights into ice premelting also have clear implications for various practical issues. They may lead to a better understanding of how concrete and other fabricated porous media degrade as a result of ice and salt growth (*9*) and how to design more durable materials; such understanding remains elusive despite hundreds of publications on freezing in porous media (*10*). Premelting and freezing in confined spaces also have considerable relevance for the cryogenic preservation of organs, the cold storage of delicate foods, and the protection of stone monuments, buildings, and art work exposed to freezing conditions.

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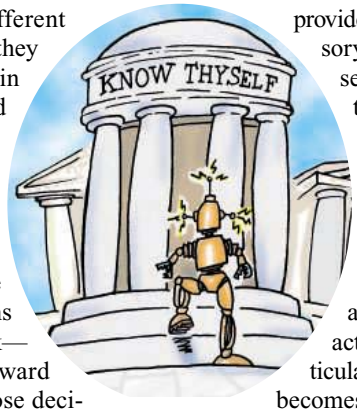
COMPUTER SCIENCE

What Do Robots Dream Of?

Robots that create and update internal models of their own structure may be able to navigate the world in a more robust way and provide a test bed for models of self-awareness.

Christoph Adami

Perhaps robots aren't so different from us after all. Like us, they need to constantly ascertain where they are in the world, and like us, they work better if they have an accurate sense of self. On page 1118 of this issue, Bongard *et al.* (*1*) show that robots equipped with an algorithm that infers their own physical structure from stored sensory data—dreams of their prior actions, so to speak—perform better in a simple forward locomotion task than robots whose decisions are not dream-inspired. Furthermore, robots that use these self-models to plan future actions can recover autonomously from injuries, by adapting their gait to compensate for the changed circumstances.



CREDIT: JOE SUTLIFF

A robot's most formidable enemy is an uncertain and changing environment. Typically, robots depend on internal maps (either provided or learned), and sensory data to orient themselves with respect to that map and to update their location. If the environment is changing or noisy, the robot has to navigate under uncertainty, and constantly update the probabilities that a particular action will achieve a particular result. The situation becomes even worse if the robot's own shape and configuration can change, that is, if its internal model becomes inaccurate. In most cases, such an event constitutes the end of that particular robot's adventure.

Bongard *et al.* aim to improve a robot's robustness in an environment that may include damage to the robot. At the beginning of a self-modeling cycle, a four-legged robot without an internal model of itself performs actions (while on a flat surface), and records its own response via tilt sensors and angle sensors in its joints. The robot then computationally tests candidate self-models, by re-imagining the actions it just performed and comparing the behavior of the model with its memory of the results—that is, the robot tries to explain the observed relationship between sensory data and leg actuation by making assumptions about its own configuration.

Even though the number of tested models is comparatively small (by only allowing a limited arrangement of limbs and their length), it is easy to imagine that many models can end up explaining the recorded behavior equally well (or equally badly). In the next stage of the cycle, the robot uses these equivalent models to find an action that would serve as the best way to discriminate among them. In other words, we could fancifully imagine the robot thinking: "Well, these three models all seem to work equally well with what I remember, but it seems to me that if I stick what I think is one of my legs out just so, then I can discover if I have a fourth leg or not." To narrow the choice of models, the robot then proceeds to test the action that provides the most information about the model's

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identity in the real world, and the cycle begins again. After 16 such cycles, the robot tests the accuracy of the final self-model by performing a set of actions that, according to this model, will result in the largest linear distance traveled, and then executing these actions. The total distance traveled can then be used as a measure of the accuracy of the robot's model of itself.

An important feature of the cycle is the active role the robot plays in determining its best self-model. Bongard *et al.* tested this feature in control experiments in which the actions taken by the robot were not informed by the self-models: For example, they forced the action synthesis algorithm to simply return a random—rather than a maximally discriminative—action. Such passive strategies fared markedly worse, as measured by the actual distance traveled after the 16 cycles. But the most dramatic difference occurred when the length of one of the robot's legs was shortened after it had gained a good sense of self. In this case, the 16-cycle algorithm was run again, this time starting with the previous best model. The active algorithm enabled the robot to adjust its gait and regain forward motion, whereas the random action controls (that is, those in which the actions were not informed by the self-model) did not.

The algorithm used by Bongard *et al.* makes use of key insights from information theory, namely, that minimizing entropy leads to maximum predictive power (*2*). A similar conclusion can be drawn for algorithms that strive to locate a robot within an unknown landscape: In this case, taking an active role in discovering the environment rather than solely relying on sensor data also leads to improved performance (*3*). Which leads us to wonder whether the approach of Bongard *et al.* could also be used to plan actions in a changing environment, based on modeling not of the self, but of the world. Active algorithms that use stochastic modeling of probabilities of beliefs (*3*) about the environment exist, but they cannot synthesize new environment models, nor generate appropriate behavior in them.

How would dream-inspired algorithms work in terra incognita? A robot would spend the day exploring part of the landscape, and perhaps be stymied by an obstacle. At night, the robot would replay its actions and infer a model of the environment. Armed with this model, it could think of—that is, synthesize—actions that would allow it to overcome the obstacle, perhaps trying out those in particular that would best allow it to understand the nature of the obstacle. Informally, then, the robot would dream up strategies for success—just as the robot constructed by Bongard *et al.* dreams about its own shape and form—and approach the morning with fresh ideas.

Although such an algorithm would require far more complex simulations than those giving rise to self-models in the work of Bongard *et al*., robots relying on this kind of navigation could play an interesting role in our quest to understand the nature of consciousness (*4*). For example, we ought to be able to record the changes in the robot's artificial brain as it establishes its beliefs and models about the world and itself, and from those infer not only its cognitive algorithms, but also witness the emergence of a personality. Thus, perhaps the discipline of experimental robot psychology is not too far off in the future. And even though the robots studied by Bongard *et al.* seem to prefer to dream about themselves rather than electric sheep, they just may have unwittingly helped us understand what dreams are for.

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CELL BIOLOGY

Sara Splits the Signal

During mitosis, signaling molecules are internalized into specialized vesicles that associate with the mitotic spindle. This ensures equal distribution into daughter cells.

Juergen A. Knoblich

Extracellular signals, such as growth factors and hormones, are received by receptors at the cell's surface and then transmitted to the nucleus via distinct cascades of intracellular signaling molecules. Because many signaling molecules are associated with intracellular membrane-bound compartments, these compartments, the signaling components, and their activation states need to be equally distributed between daughter cells during cell division. This is particularly important in developing tissues, where morphogens can elicit concentration-dependent responses at very long ranges, and even small variations in their concentration can create very different effects. On page 1135 of this issue, Bökel *et al.* show that cells in the developing wing of the fly *Drosophila melanogaster* contain a specialized subset of intracellular vesicles called Sara endosomes, whose main function seems to be equally distributing components of the transforming growth factor–β (TGF-β) signaling pathway during cell division (*1*). This mechanism ensures that the activation state of the signaling pathway remains precisely the same in both daughter cells.

The effects of the morphogen TGF-β on vertebrate and invertebrate tissue development rely on a relatively simple pathway (see the figure). TGF-β binds to two cell surface proteins called type I and II receptors and induces their dimerization. The type II receptor phosphorylates and activates the type I receptor, which in turn phosphorylates the transcription factor R-Smad. Phosphorylated R-Smad binds to a co-Smad to form an active transcription factor that translocates into the nucleus and induces the expression of target genes.

Sara (Smad anchor for receptor activation), a conserved, membrane-associated adaptor protein, simultaneously binds to the TGF-β–receptor complex and the R-Smad (Mad in *Drosophila*) (*2*). Sara contains a so-called FYVE domain that binds phosphatidylinositol 3-phosphate [PI(3)P], a membrane phospholipid that is primarily found on early endosomes. Although earlier work suggests that Sara is required for TGF-β signaling and recruits the receptor–R-Smad complex into endocytic vesicles, subsequent reports have led to conflicting views on the precise function of Sara. Sara can also bind to the phosphatase PP1c—a negative regulator of TGF-β signaling—and therefore could also inhibit TGF-β signaling (*3*). Furthermore, experiments addressing the role of endocytosis in TGF-β signaling have given conflicting

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results (*4*–*7*). Endocytosis is not required for the Sara–TGF-β receptor–R-Smad complex to form (*4*), so it is not clear how endocytosis would activate TGF-β signaling.

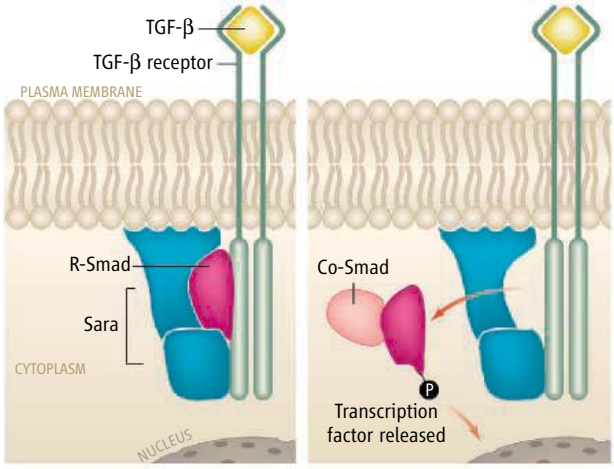
To analyze the function of Sara, Bökel *et al*. used development of the *Drosophila* wing as a model system for TGF-β signaling. Wing cells recognize their position along the anterior-posterior body axis from the local concentration of Dpp (Decapentaplegic), one of several TGF-β–like molecules found in *Drosophila*. Dpp is secreted from a narrow stripe of cells in the center of the developing wing and forms a concentration gradient that turns on target genes in other wing cells in a concentration-dependent manner. Previous experiments have suggested that Dpp might spread across the developing wing by transcytosis: It binds to its receptor, Thickveins, on the cell surface, is internalized, and is subsequently secreted back into the intercellular space through the process of transcytosis. Because a fraction of the receptor-ligand complex is degraded during each round of transcytosis, this could lead to the formation of a stable concentration gradient of Dpp across many cell diameters.

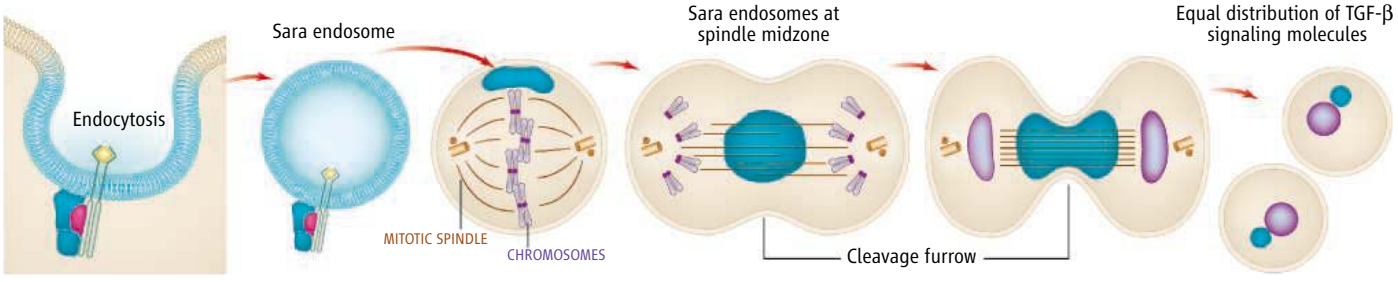
Bökel *et al*. observed that loss of Sara causes only mild defects during wing development. Wing veins were occasionally branched or duplicated, but the overall Dpp-related patterning of the wing was unaffected. On a cellular level, however, the defects were dramatic. In wings of wild-type flies, the activity of the Dpp pathway was highly similar in sibling cells, as indicated by similar amounts of activated Mad. In contrast, wing cells of mutant flies lacking Sara show a broad range of differences, with many sibling cells differing by more than a factor of 2 in the amount of active Mad. Surprisingly, however, the overall gradient in receptor activity was normal in the mutant flies. Bökel *et al*. attribute this to the selective death (apoptosis) of cells with abnormally high levels of TGF-β signaling that they observe in Sara mutants. Selective elimination of cells with abnormal levels of Dpp signaling has been described before (*8*), although in this case, apoptosis occurred in cells with lower signaling levels. Thus, Sara is not generally required for Dpp signaling but is involved in the proper distribution of the Dpp signal during mitosis.

How might Sara perform this function? Sara is localized on a specific subset of early endosomes, and Bökel *et al.* used immunoelectron microscopy studies to show that it concentrates on the limiting (outer) membranes of multivesicular endosomes. In interphase of the cell division cycle, these Sara endosomes concentrate apically. In anaphase, however, they associate with the spindle midzone, and during cytokinesis they split into two distinct compartments that associate with either end of the central spindle and are segregated into the two daughter cells. Bökel *et al.* demonstrated that the TGF-β receptor Thickveins is concentrated in Sara endosomes during mitosis. In Sara mutants, these endosomes still form. However, they no longer contain Thickveins, and this may explain the differences in TGF-β signaling between the two daughter cells. Random distribution of endocytic vesicles into the daughter cells is imprecise (*9*), and the specialized mitotic morphology of Sara endosomes seems to ensure the equal distribution of their contents into the two daughter cells.

Is the Sara endosome a peculiarity of *Drosophila*? In vertebrate cells, some endocytic vesicles associate with the spindle midzone during late mitosis and are involved in cytokinesis (*10*). Although these vesicles have not yet been implied in signaling, vertebrate cells seem to have the principal ability to create spindle-associated endosomes that resemble the Sara endosomes, at least in their morphology. Whether they are involved in TGF-β signaling could be tested because their formation requires the small G protein ARF-6, whose expression or function could be blocked.

A distinct distribution during mitosis was also shown for *Drosophila* recycling endosomes (those vesicles that traffic back to the plasma membrane). During symmetric cell division, recycling endosomes concentrate around the centrosomes during anaphase and telophase (*11*) and behave identically in both daughter cells. During asymmetric division, however, endosome recycling can be suppressed in one of the daughter cells to generate differences in protein trafficking and signaling. This results in different cell fates of the two daughter cells (*12*). It will be exciting to analyze whether the Sara endosome is also used in certain biological contexts to distribute TGF-β signaling unequally between daughter cells. In any case, the new study shows that the analysis of vesicular trafficking in the context of a whole animal can reveal features that might never have been discovered in cultured cells.





Daughter cells are treated equally. Sara simultaneously binds to membrane phospholipids, the TGF–β–receptor complex, and R-Smad. Upon receptor activation, R-Smad is phosphorylated and released from Sara to bind the co-Smad and form a functional transcription factor. During cell division, Sara endosomes and their contents are recruited to the mitotic spindle midzone and are equally distributed into the two daughter cells.

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CHEMISTRY

Breaking the H_2 Marriage and Reuniting the Couple

A nonmetal compound enables the reversible splitting of dihydrogen. Related compounds may find use in chemical transformations and in hydrogen activation or storage.

Gregory J. Kubas

Molecular hydrogen (H_2) is not only a valuable future fuel; it is widely used today in chemical reactions, such as the addition of H_2 to organic compounds. These "hydrogenation" processes are among the largest-volume human-made chemical reactions: All crude oil is treated with H_2, and 10^{10} tons of ammonia fertilizer are produced annually via catalytic hydrogenation. Any small improvement in catalyst efficiency, cost, or availability would help to cut the cost of these important processes.

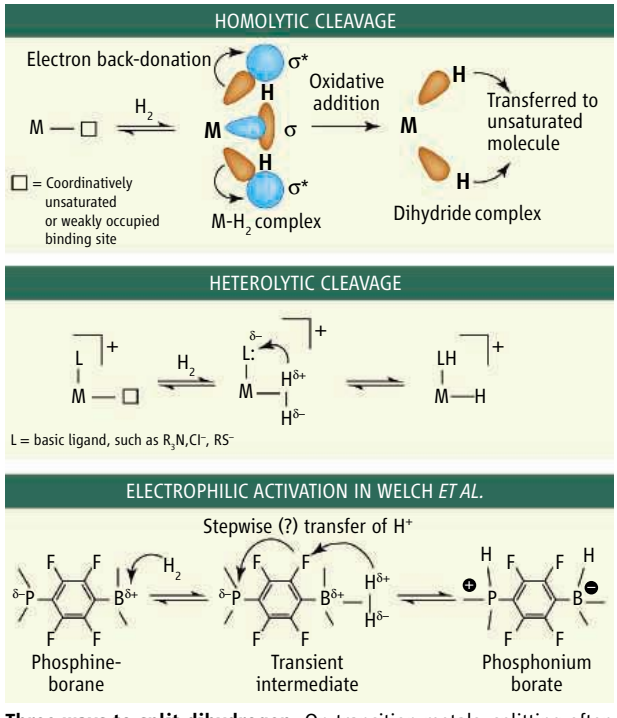
But such improvements are hard to come by. As in a strong marriage, the H_2 molecule is held together by a tight bond. It can be split apart in a controlled manner on metal catalysts and a few nonmetal compounds. But reuniting the couple—that is, reversing the process to reform the H–H bond—is more difficult and has never been accomplished on a nonmetal compound. Welch *et al*. have now accomplished this feat, as described on page 1124 of this issue (*1*).

This exciting finding is important not only for chemical reactions involving hydrogen, such as catalytic hydrogenations, but also for hydrogen-fuel storage and production. The metal-free aspect is highly relevant, because the precious metals such as platinum that are widely used in catalysis can be environmentally unfriendly, as well as costly or in short supply. Also, main-group compounds are less heavy, which is important for hydrogen storage.

On metal complexes and enzymes, the H–H bond-splitting process—often referred to as H_2 activation—requires the separation of the two electrons in the H–H bond to form metal-hydride complexes (see the figure, top panel). However, the molecular mechanism by which the H–H union breaks up has been difficult to establish, because H_2 molecules cannot easily be caught in the act of binding to a metal or other third party (usually the first step in weakening and eventually breaking up a marriage).

The discovery in 1984 of a nearly intact H_2 molecule coordinated to a metal complex did catch this act in intimate detail (*2*). In the complex, the H_2 binds side-on to the metal, primarily via donation of the two hydrogen σ electrons to a vacant metal d orbital. The resulting dihydrogen complex has a stretched H–H bond (see the figure, top panel). This ménage à trois has now been seen in hundreds of transition metal complexes.

The binding to the metal in these complexes is relatively weak and can be highly reversible: H_2 can be added and removed by mild pressure and/or temperature variation. The binding is stabilized by back-donation of electrons from a filled d orbital to the antibonding orbital (σ*) of the bound H_2 (*3*, *4*). Increasing the back-donation to σ* (for example, by ligand variation) causes the H-H bond to break to form two M–H bonds (see the figure, top panel); the latter can then transfer hydrogen atoms to organic or other substrates. This is termed homolytic bond cleavage.



Three ways to split dihydrogen. On transition metals, splitting often proceeds via homolytic cleavage [**(top)**; also referred to as oxidative addition]. Brown electron orbitals are filled; blue orbitals are vacant. A second mechanism, heterolytic cleavage **(middle)**, has been observed for transition-metal systems. The heterolytic, reversible cleavage of dihydrogen on a phosphine-borane reported by Welch *et al.* is likely to proceed via a third, related mechanism called electrophilic activation **(bottom)**.

H_2 can also bind to strongly electrophilic nonmetals via electron donation. An example is CH_5^+, which is now viewed as an H_2 molecule bound to CH_3^+ in a highly dynamic fashion (*5*). However, this type of compound is much less stable than the metal-H_2 complexes described above. Also, homolytic H–H bond breaking is not possible here, because such main-group atoms cannot back-donate electrons.

How does the main-group compound reported by Welch *et al.* cleave and reversibly reform H_2? It must be by heterolytic cleavage of H_2, a second, more versatile pathway in catalytic hydrogena-

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tion (*6*–*8*). In this case, the union in H_2 becomes polarized upon interaction with a positively charged or electron-poor (electrophilic) metal, resulting in splitting to a metal hydride and a proton (H^+) that can easily transfer to a ligand (see the figure, middle panel) or external substrate. H_2 can become more acidic than sulfuric acid when bound to an electrophilic metal center (*7*).

Heterolytic cleavage has been mainly observed in transition-metal complexes, most notably as part of the mechanism of Noyori's asymmetric hydrogenation systems (*9*) and hydrogen activation in hydrogenase enzymes (*10*). However, it can also occur on nonmetal centers. For example, metal-bound sulfide ligands are known to react with H_2 to form SH ligands (*11*), and metal-free hydrogenation of ketones on strong bases appears to proceed via base-assisted heterolysis of H_2 (*12*, *13*).

The phosphine-borane species used by Welch *et al.* combines a strongly electrophilic center (boron) with a nearby nucleophilic site (phosphorus) that can apparently accept the proton from heterolytic splitting of H_2. Several mechanisms are possible, but it is likely that H_2 initially interacts with the electrophilic boron center, followed by proton migration from an H_2-like complex to the basic phosphorus atom, which is separated from the boron center by a perfluorophenyl linker (see the figure, bottom panel). This migration could proceed stepwise via the linker, as proposed by Welch *et al.*, or could be assisted by the solvent.

Regardless of the mechanism, the discovery is important because of the reversible nature of the hydrogen activation. Materials for hydrogen storage are a vexing challenge, particularly for vehicles, because energetically favorable extrusion of hydrogen from materials is rare [a recent example is H_2 evolution from hydride-like organic compounds (*14*)]. It can also be very difficult to add hydrogen back. Furthermore, the materials must be lightweight, reducing the prospects for known, easily reversible systems such as dihydrogen or hydride binding to transition metals. Amine borane is a popular candidate and also combines both electrophilic (B) and nucleophilic (N) centers. However, these centers are directly bonded, whereas in the phosphine-borane described by Welch *et al*., they are separated by a linker, increasing the electrophilicity of B and the nucleophilicity of P.

The hunt is now on for compounds similar to that reported by Welch *et al.* for hydrogen storage and activation. Biomimetic hydrogen production by splitting of water, particularly in processes that use sunlight, is also a challenge and may take a cue from models of the iron-containing active site of hydrogenases (*15*). Here, the mechanistic reverse of heterolytic splitting of H_2 will be crucial, involving formation of hydrogen from protons and electrons, a highly reversible rapid process in hydrogenases. Clearly, there are now many new avenues for chemical bond splitting and transformations.

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BIOCHEMISTRY

RNA Polymerase, a Scrunching Machine

Jeffrey W. Roberts

RNA polymerase stores energy to break its initial bonds with DNA by scrunching the single strands of DNA that were unwound in the region where the polymerase started RNA synthesis.

RNA polymerase (RNAP) mediates the critical steps in gene expression and is thus an important target for mechanistic analysis by sophisticated biophysical techniques. A striking example is the subject of two reports in this issue of *Science*. On pages 1144 and 1139, Kapanidis *et al.* and Revyakin *et al.* (*1*, *2*) illuminate the initial steps of making an RNA chain by showing how the energy of nucleoside triphosphate hydrolysis is captured to break the enzyme loose from its tight contact with DNA at the beginning of the transcribed segment. The results reveal an unexpected structure of DNA in the transcribing complex that may well have an important role in regulating gene expression.

Attempts to explain a strange property of RNAP stimulated these experiments. Through the process of transcription, coiled and double-stranded DNA is unwound into single strands, and RNAP synthesizes RNA that is complementary to the templating strand of DNA. Instead of continuing every RNA chain that it starts, RNAP tends to falter badly, releasing most chains near the beginning of transcription—generally after 5 to 10 nucleotides have been assembled into RNA—and then starting over, a process called "abortive initiation" (*3*–*6*).

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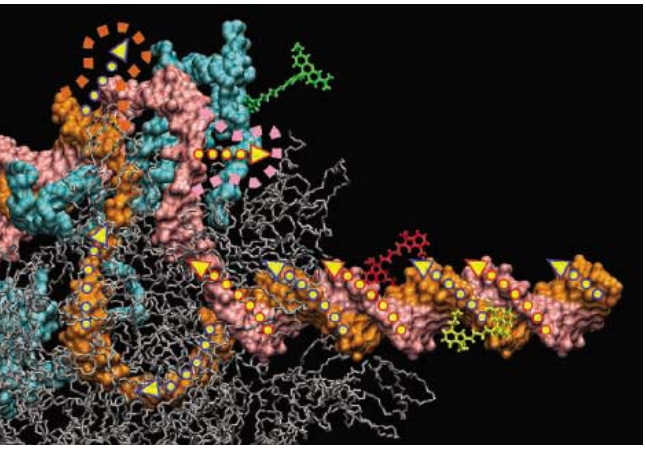
It has long been clear that the cause of abortive initiation is the failure of RNAP to break the bonds that bound it to DNA—specifically, to a region called the promoter—in the first place. For bacterial RNAP, these bonds connect sigma factor regions 2 and 4 (in the example of the σ^{70} RNAP) to the promoter –10 and –35 elements, respectively. Comparable but more complex networks of initiation factors presumably bind eukaryotic RNA polymerase II to its promoter. Nearly 20 years ago, Straney and Crothers (*7*) suggested that energy to break promoter contacts is stored in a "stressed intermediate" form of the RNAP-promoter complex during the first few nucleotide addition steps in RNA synthesis and that abortive cycling represents failed attempts to use this energy productively.

But how might the energy be stored? One structure-based proposal of Darst and associates (*8*) suggests that emerging RNA must actively force the protein linker between sigma domains 3 and 4 from the RNA exit channel of RNAP. This model is specific in detail to bacterial polymerase but possibly applicable to other RNAPs.

However, a more general answer comes from the two new reports, designed to answer a related question: How can the transcribing complex be flexible enough to allow synthesis of RNA 10 nucleotides long (or more) and at the same time keep its grip on the promoter? Clearly, something has to bend or move, and

the two reports consider three possibilities. First, RNAP itself might be flexible, allowing the active center to move downstream along DNA with the bubble of "melted," unwound DNA where templating of RNA occurs, whereas sigma (in the case of bacterial RNAP) stays bound to the promoter elements. In this scenario, energy is stored in the distorted protein. Second, promoter-sigma bonds might break transiently, allowing RNAP to move downstream during RNA synthesis but diffuse back to the promoter when the abortive RNA is released. Third, the DNA bubble might unwind downstream DNA without any movement at the upper end, meaning that sigma stays bound to the promoter and melted downstream single-stranded DNA is "scrunched" into a nearly rigid enzyme. Energy would then be stored as melted DNA. Scrunching of DNA in fact was detected originally in an initiation complex of the single subunit T7 RNA polymerase revealed through atomic crystallography by Cheetham and Steitz (*9*).



Ready to scrunch. The model shows an open promoter complex of bacterial RNA polymerase (RNAP) poised to begin RNA synthesis, with arrows designating motions of DNA segments that occur as the first nucleotides are combined to form an RNA chain. Downstream DNA (on the right) rotates inward and separates in the active site channel of the polymerase. Template (orange) and nontemplate (pink) DNA strands follow the indicated paths, moving as the RNA chain (not shown) is polymerized on the template DNA strand. Sites of extrusion of single-stranded DNA are shown by outlined arrows at the top. σ^{70} (blue); RNAP core chains (gray); a fluorescent tag on σ^{70} region 2 used for FRET (green); a fluorescent tag on downstream DNA (initial position in yellow; scrunched position in red).

Two methods of single-molecule analysis were used to distinguish these models, and these methods give complementary results. Single-molecule fluorescence resonance energy transfer (FRET) was used to measure relative movements of various reference points on the transcription complex during transcription, specifically sites on RNAP near its upstream and downstream edges, and sites in upstream and downstream DNA. The result is that downstream DNA moves closer to the other sites during abortive initiation, but upstream DNA and sites within RNAP do not move relative to each other. This result is predicted only by the scrunching model and rules out either protein flexibility or RNAP excursions from the promoter during abortive cycling.

In the second method, DNA unwinding induced by RNAP was measured by nanomanipulation of an extended template DNA on which melting can be measured, to 1-base pair resolution, as a change in supercoiling-induced contraction. The result showed the expected melting when the initial promoter bubble (open complex) formed, with further melting corresponding to the length of the abortive RNA that was made—as predicted by the scrunching model. Furthermore, the nanomanipulation method showed that promoter escape is preceded by scrunching in most if not all transcription events, considering the 1-s limit of detection. The authors conclude that scrunching is an obligatory step in promoter escape, which occurs as scrunched DNA rewinds and exits through the back of the enzyme, breaking the bonds to promoter sequences.

This discovery has implications for several basic processes of transcription and thus of gene expression. Most strikingly, the scrunching model implies a novel and unexpected structure. Scrunched single-stranded DNA eventually would be extruded from the enzyme channel at predictable sites (see the figure) and thus would be available for interaction with transcription regulatory proteins. No such interactions are yet known, but it seems unlikely that this opportunity has been wasted in evolution. These experiments also define the critical structure present when the choice between elongation of RNA and abortive loss of RNA is made—a mysterious process and potential point of regulation. Knowledge of the structure of the abortive complex and the stability of the scrunched state should guide studies of the mechanism of bacterial Gre proteins (and their eukaryotic counterpart TFIIS), which inhibit abortive initiation in vitro and are likely to be important modulatory elements of both initiation and elongation of RNA synthesis in the cell.

Scrunching also could explain a perplexing incongruity between transcription initiation in higher eukaryotes and the otherwise closely similar model, the yeast *Saccharomyces cerevisiae*. In the former, initiation occurs at a fixed distance from a dominant site of polymerase interaction in the promoter called the TATA box. In yeast, initiation occurs at variable and more distant sites. The latter could be explained if the melting function of the initiation factor TFIIH used adenosine triphosphate energy to pump DNA into a scrunched state as the enzyme searched for a suitable initiation site on the template DNA strand (*10*). In this case, many nucleotides of melted DNA could be extruded out the sides of the polymerase (see the figure).

Equally interesting is the implication of scrunching for the structure of transcription complexes paused during elongation. Paused transcription elongation complexes are important sites of regulation, and certain examples are likely to be similar to abortive complexes at the promoter. At such sites, RNAP constrained from upstream could continue transcription for some distance by drawing in downstream DNA. One example occurs in an antitermination regulatory circuit of the *Escherichia coli* bacteriophage lambda in which the sigma initiation factor rebinds a promoter-like sequence in downstream DNA, forming a specialized elongation complex that is substrate for an antitermination protein (*11*). This structure is almost certainly formed by scrunching, and a stable scrunched state could be the regulatory target. Promoter-proximal paused transcription complexes are an important feature of eukaryotic transcription, well characterized for the heat-shock promoters in *Drosophila melanogaster* and the human HIV promoters and conjectured to occur widely (*12*); DNA scrunching could well be an element of their formation and regulation. It is a good bet that scrunched DNA will appear in future detailed views of transcription regulatory complexes.

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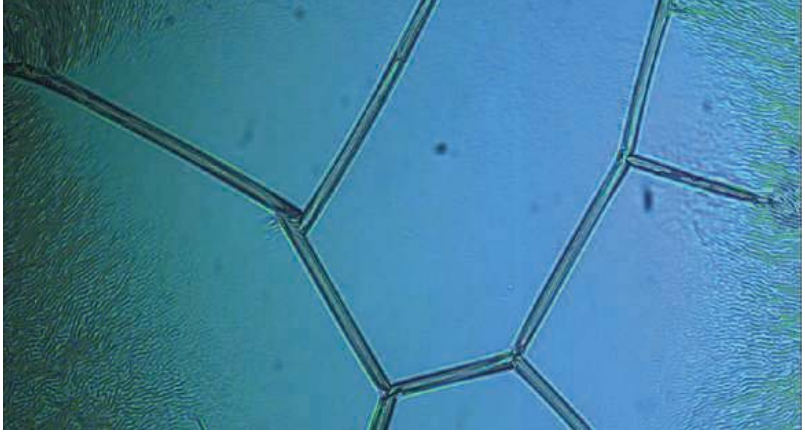

INTRODUCTION

The Right Combination

IN THE SIMPLEST SENSE, A COMPOSITE IS AN OBJECT MADE UP OF TWO or more distinct parts. A common example is a composite image that may come from a series of photographs that together tell a bigger story than could be achieved with only one image. Within materials science, composite materials are put together from two or more components that remain distinct or separate within the final product. They can be as simple as a matrix material that envelops a reinforcing material, as when concrete surrounds steel bars that help prevent the concrete from failing under tension. Beyond this simple construct, composite forms now include layered structures and reinforcing agents that act in all three dimensions. The challenge is that the options for making a composite are almost limitless, but only a few combinations of materials will combine synergistically, and the design criteria may not be obvious. To design a material that will absorb more energy before breaking, a weaker reinforcing material may be added. When this composite fails, it may form a much larger number of cracks, and it is the additional crack length that makes the material tougher.

This special section examines contemporary composite materials from three different perspectives. Hogg (p. 1100) describes their very practical and important use in crafting energy-absorbing armor to protect vehicles and people. This application demands a remarkably broad range of properties, in that stopping a knife attack is very different from stopping a bullet; and in the case of projectiles, stopping the second bullet or mortar is as important as stopping the first.

Imagine a single combination of matrix and reinforcing fiber. Aside from the weight fraction of fibers used, variables include the fibers' distribution pattern in the matrix, their orientation, and their length profile, among other characteristics. Every permutation may respond differently to the range of repeated stresses that, for example, an airplane must withstand. The challenge of certification in this context is a major impediment to the widespread use of composites in critical applications. However, the tide may be turning. Cox and Yang (p. 1102) review the improving accuracy of models and simulations to make it possible to accurately extrapolate the failure properties of a range of composites from a limited data set.

Balazs *et al.* (p. 1107) focus on the challenges of designing new composites. They offer a detailed look at what happens when inorganic nanoparticles are embedded in a polymer matrix. Because of the comparable dimensions of the components, both enthalpic and entropic effects come into play.

As the design of new materials becomes more costly and complex, the synergistic fusion of existing materials into a better composite becomes an increasingly attractive fabrication strategy. It is hoped that the perspectives presented here may encourage new thinking toward bringing diverse materials and scientists together to make better composites.

– MARC LAVINE

CREDIT: BALAZS *ET AL.*

Materials Science: Composites

CONTENTS

Perspective

1100 Composites in Armor
P. J. Hogg

Reviews

1102 In Quest of Virtual Tests for Structural Composites
B. Cox and Q. Yang

1107 Nanoparticle Polymer Composites: Where Two Small Worlds Meet
A. C. Balazs, T. Emrick, T. P. Russell

See also related ScienceCareers material on page 1043

Science

PERSPECTIVES

Composites in Armor

Paul J. Hogg

Composite materials are traditionally regarded as materials that can save energy in large structures associated with transport. They are used to produce lightweight structures for fuel-efficient aircraft such as the new Boeing 787 Dreamliner; lightweight cars from Lotus, Ferrari and TVR; and high-speed trains, speedboats, and racing yachts. Now, however, some of the most interesting applications of composites are those where the materials are used to save lives and protect property by absorbing the energy of projectiles, impacts, and crashes.

We have become used to the sight of Formula 1 cars hurtling into obstacles and suffering horrendous crashes, only for the driver to emerge unscathed if a little shaken. The secret of this remarkable ability to protect the drivers lies in the wholesale use of carbon-fiber composites for the cars' structure and the exploitation of that material's ability to fragment in a controlled way. Through fragmentation, a composite absorbs more energy than conventional engineering materials per unit of weight. Most conventional engineering materials such as metals absorb energy in a crash by plastic deformation during the crumpling of the structure. Composites, on the other hand, can absorb energy by a process of microfragmentation. Most industrial composites are produced from brittle fibers and a brittle polymer matrix, which is not an encouraging start for developing materials that absorb a lot of energy on fracture. However, in a composite, the presence of countless interfaces between the constituents provides opportunities for multiple cracking, and the cumulative effect of myriads of tiny brittle fracture events is to create an ability to absorb energy that can surpass that of monolithic ductile materials. Composites based on organic fibers, such as Kevlar (polyaramid) and Spectra (ultra-high-molecular-weight polyethylene), can also absorb energy by deformation of the fiber itself. Composite structures that absorb energy in crashes by crushing are now widely used in helicopters and trains as well as Formula 1 cars. They are even slowly becoming adopted for use in more conventional automobiles, albeit the rather expensive ones such as Aston Martins.

This ability to absorb energy is also being widely exploited in the development of dedicated safety systems that are used variously for the protection of structures, property, and people. The casings of jet engines are being produced from fiber-reinforced composites to better protect them from broken fan blade debris and to protect the aircraft cabin from catastrophic damage if the blades break in service. Similarly, the undershields of many cars are produced from fiber composites to protect delicate parts of the car against stone damage on rough terrain. More interestingly perhaps, composite armor is now protecting military vehicles and providing the basis for most forms of personal body armor. The advent of new materials, including nanomaterials, is providing many new options for the materials designer to use to create ever-more-exotic composites to meet the increasing threats posed to people and property.

Providing armor for military vehicles used to be a relatively simple task. The basic concept was that in order to slow down and render harmless a projectile, a large mass was put between the vulnerable parts of the vehicle and the projectile's likely trajectory. The conservation of momentum did the rest. This approach relied on the armor and its material being strong enough not to break before the projectile had stopped. Steel was an admirable choice for such duties; however, steel is heavy and increasing the level of steel protection to meet increasing threat levels inevitably increases the weight of a vehicle dramatically.

The ongoing need for a lightweight structure to allow the military vehicle to move quickly and to be transported by air has led the push toward new materials. Composite armor based just on fiber composites has been widely used to provide additional protection in light vehicles such as Jeeps and Land Rovers. Here the panels are typically based on S-2 glass fibers [composition: SiO_2 65 weight % (wt %), Al_2O_3 25 wt %, and MgO 10 wt %], which have a higher strain to failure and modulus than conventional low-alkali, aluminoborosilicate E-glass fibers (composition nominally SiO_2 54 wt %, Al_2O_3 14 wt %, CaO + MgO 22 wt %, and B_2O_3 10 wt %). Carbon fibers are not used in such applications because the energy absorbed is less for a given weight. This is because the strain to failure of the carbon fiber is lower than that of glass fiber, and the carbon fiber panel does not exhibit as high a level of microcracking before final penetration as a glass fiber composite. Although glass fiber composites are effective against small-arms fire, they are inadequate for more serious threat levels posed by large-caliber munitions and armor-piercing rounds. In developing protective armor to meet such threats, different types of composites are now being explored, including laminates of different materials which may themselves be fiber-reinforced composites or monolithic systems such as ceramics and metals.

Integral armor is a term used to describe a laminated material consisting of a front-facing layer whose purpose is solely to blunt and abrade the incoming round. Meanwhile, a second layer supports the facing material during this initial impact and then deforms and absorbs energy. This combination can defeat an armor-piercing round. Typically, the front-facing material is a ceramic, usually Al_2O_3, although other materials including TiB_2 have been considered. A hard and dense ceramic is essential because the material effectively pulverizes into tiny fragments ahead of the projectile, which is worn down and abraded as it passes through the layer. Originally the backing layers were light metal such as aluminum, but now a fiber composite will provide the best combination of energy absorption (stopping power) and weight.

Developments in this field have all been driven by the many different demands that are being put onto the materials in various applications. A multi-hit capability requires the ceramic to be in the form of small tiles, so that the shattering of the outer tile affects only a small zone. The use of intermediate layers between the ceramics and the fiber-composite backing layers is valuable in reducing the spread of damage. Some new concepts include using a peg type of ceramic, almost like a short bolt and a hexagonal head that connect together on the surface and slot into holes in a backing structure. Probably the lightest armor on the market is supplied by IBD in Germany (*1*), which now uses a nanocrystalline ceramic tile to minimize the spread of damage. Improved ceramic properties are being claimed for systems manufactured using a pressureless sintering technique, followed by hot isostatic pressure for consolidation to achieve a very dense >99% ceramic material. Other novel materials being used for the ceramic tile include functionally graded Ti-TiB2 systems (*2*). The performance of the ceramic itself is subject to modification by physical constraints. A very thin fiberglass polymer composite bonded onto the front face of a ceramic tile will improve the ballistic performance by constraining the channel through which the plume of debris is ejected against the advancing projectile (*3*).

In addition to materials developments, the combination of new material forms in a composite armor is proving interesting. Foamed metal layers have been proposed in the United States as intermediate layers between the ce-

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ramic and the fiber composite in order to reduce stress-wave transfer and to allow the projectile to be slowed by crushing the foam before the fiber composite is deformed (*4*). This will also reduce the extent of intrusion by the deformed armor into the space on the reverse face from the impact. This may be may be important if space is limited and critical if the armor rests against sensitive equipment or a person, as in the case of helmets.

One of the most important drivers is the reevaluation of the role of armor itself in military vehicle design. In the United Kingdom, DERA (now QinetiQ) successfully produced a lightweight glass-fiber composite hull for their Advanced Composite Armoured Vehicle Platform in order to reduce weight (*5*). However, by the time the external armor, turret, running gear, and gun were added, the total weight saving of 4 tons was considered small compared to the original steel vehicle's weight of 28 tons. The concept of hanging armor off a basic vehicle hull is now changing to the idea of integrating the armor into the main structure in order to reduce weight to a greater degree. This might provide a greater impetus to adopt carbon-fiber composites, which will provide the lightweight structure, and if thick enough, can absorb a respectable amount of energy. Hybridization of the carbon fiber with less-expensive and more-ductile fibers may allow greater ballistic protection and reduce costs; some promising materials such as RINO, based on combinations of carbon/nylon fibers developed by Cytec Industries/Matrice Materials Systems, are now being evaluated. If an integral armor solution is adopted, then the composite structure must also retain adequate postballistic impact strength to allow the vehicle to complete its mission, and this may change the optimum combination of materials in any given situation. The use of complex three-dimensional (3D) textiles as the fiber form, as opposed to conventional 2D fabrics, is likely to be part of any solution that requires good postballistic impact properties from the composite armor. The introduction of more complex fiber architectures will, however, complicate the already difficult modeling task of predicting properties ahead of experimental evaluation. The difficulty of adequately modeling the ballistic behavior of composite materials is currently a major constraint in their application.

The major developments in fibers themselves for composites that have a bearing on armor are the extremely tough and stiff rigid-rod organic fibers such as *p*-configured poly (*p*-phenylenebenzobis-oxazole (PBO) (*6*) and poly-{2,6-diimidazo[4,5-b:4′,5′-e]pyridinylene-1,4(2,5-dihydroxy) phenylene} (PIPD, known in fiber form as M5), that have been developed in recent years (*7*). These materials are more suited to flexible armor as opposed to the rigid armor used in tanks. These fibers must be able to extend to a larger extent than glass or carbon fibers in order to get the maximum benefit from their inherent toughness, and this is restricted in the rigid panels developed for structural armor by the low strain to failure of the polymer matrices used.

Flexible composite armor, mainly used for body protection, is based on a different concept. The material is principally composed of high-stiffness organic fibers in a dense fabric stack. The stiffness of the fiber ensures that the load is distributed over a large area of the material, and penetration of the fabric requires stretching of the fibers, which absorbs a lot of energy. Flexible armor introduces another design complication in that the system must absorb the energy of the projectile, but cannot be allowed to deform so extensively that the wearer of the armor is crushed in the process. Aramids such as Kevlar and Twaron and aliphatic polymers such as Spectra and Dyneema have dominated this field. The new M5 and PBO fibers have much higher moduli, greater than those of many carbon fibers, coupled with very high strengths in the region of 5 GPa. There has, however, been concern regarding the degradation suffered by PBO fibers because of the effects of moisture, which has alarmed potential users. Nevertheless, U.S. Army tests suggest that M5 fibers will provide weight savings of 40 to 60% for protection equivalent to that provided by Kevlar fibers (*8*). The weave of the fabrics has a bearing on the ballistic protection, but another important effect is the friction loading between the fibers and fiber bundles. Recognition of this has led to astonishing new developments in what has been described as liquid armor.

In these systems, the ability of a liquid-like system to stiffen rapidly under a shear stress is used to produce an instant "solid." Various groups have studied this phenomenon using nanoscale silica particles thickening liquids such as water (*9*) and ethylene glycol. Rapid loading make the system stiff and strong. When these liquids are used to impregnate ballistic fabrics, during the shock loading by a projectile, the flexible fabric becomes surrounded by an instant stiff matrix, forming a composite and providing ballistic protection. The improvement of the fabrics in terms of the ballistic limits can be doubled by this shear thickening action of the surrounding liquids. The U.S. Army is extremely interested in this concept, which could find use in other personal protection applications, notably in sporting goods.

It cannot be long before more types of novel nanomaterials are being used as parts of more complicated composite armors. Carbon nanotubes exhibit extremely high strength and stiffness, whereas inorganic fullerenes offer an ability to absorb energy (*10*), as do polymer matrices with microarchitectures (such as extendable microframes based on epoxy systems) (*11*). These materials could be incorporated into armor panels to reduce weight if the costs are not prohibitive. The personal armor area looks set to expand, and some novel approaches to protecting against both bullet and knife attacks are being developed. For a knife attack, it is desirable to provide an additional outer layer of a hard material, usually in tile form, to blunt the weapon and to stop the reinforcing fibers being cut. Novel developments in this area are based not so much on the materials, which may be steel, titanium, or ceramic, but on the arrangement and organization of the tiles. A "dragonskin" system developed in the United States has been patented, which consists of milled disks of a hard material that provide a flexible layer with constant thickness attached to a backing layer of ballistic fabrics.

Although there is an exciting raft of new materials on offer for armor development, the design of armor systems and the selection of materials currently rely to a great extent on on experience, empiricism, and intuition. The analytical and numerical modeling tools available are not yet able to fully identify optimum properties of fibers, fabrics, and structures to provide an armor system best tailored to meet a specific threat. Recent advances in modeling are, however, encouraging and look likely to provide a better description of the fracture processes taking place during a ballistic impact (*12*). More accurate models will be able to guide the materials engineer to both develop and use materials more effectively. The only restriction on the growth of composites for protective uses will then be the limits of the imagination of the composite engineer. Thankfully, we seem nowhere near reaching that limit for some time to come.

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REVIEW

In Quest of Virtual Tests for Structural Composites

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The difficult challenge of simulating diffuse and complex fracture patterns in tough structural composites is at last beginning to yield to conceptual and computational advances in fracture modeling. Contributing successes include the refinement of cohesive models of fracture and the formulation of hybrid stress-strain and traction-displacement models that combine continuum (spatially averaged) and discrete damage representations in a single calculation. Emerging hierarchical formulations add the potential of tracing the damage mechanisms down through all scales to the atomic. As the models near the fidelity required for their use as virtual experiments, opportunities arise for reducing the number of costly tests needed to certify safety and extending the design space to include material configurations that are too complex to certify by purely empirical methods.

Predicting the failure of materials is one of the oldest problems of engineering (*1*) and one of the least perfectly solved. Even in modern times, after a century of the formal study of fracture mechanics (*2*, *3*) and the advent of computational stress analysis, predictions remain closely tied to empirical data gathering. In the case of structures upon whose integrity human lives depend, the burden of testing to prove safety is immense: A typical large airframe, for example, currently requires ~10^4 tests of material specimens, along with tests of components and structures up to entire tails, wing boxes, and fuselages, to achieve safety certification (*4*). Although stress analysis is an excellent tool for predicting the distribution of loads throughout the structure when its behavior is linear-elastic, once damage begins, prediction becomes problematic. The fundamental difficulty is that damage in tough engineering materials (excluding brittle materials such as glass) involves extremely complicated nonlinear processes acting from the atomic scale (e.g., dislocations and bond rupture) through the microscale (e.g., microcracking, crazing in polymers, plasticity in metals) and on up to the scale of the structure itself (e.g., large cracks and buckling modes).

Our shortcomings in tackling such a problem are not merely in computational power; the more decisive challenge is to categorize and characterize the many possible mechanisms of damage and build them into a model in a realistic way. Even in a mainly empirical approach to predicting failure, great care must be taken to assure that tests are conducted that will reveal all mechanisms that might appear in service. The history of disasters is replete with cases where the critical mechanism eluded the test regimen. When prediction is taken up as a theoretical challenge, we need not only to know all the mechanisms but also to have a model for each that will correctly represent its effect on the progression to failure.

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Bottom-Up and Top-Down Models

Current research devolves broadly into two approaches to failure prediction, the bottom-up and the top-down methods. The bottom-up method, which from the first era of fracture and dislocation theory in the mid-20th century has attracted as many physicists as engineers [e.g., (*5*)], seeks to simulate failure by building up detailed models of atomic and molecular processes by means of quantum mechanics and classical molecular dynamics. Processes modeled with increasing realism include crystal plasticity (*6*–*8*), bond rupture in relatively brittle materials for simple crack geometries (*9*–*11*), and the deformation of collagen molecules and fibrils in natural composites (*12*, *13*). The difficulty with the bottom-up method is that the intervals of time and the size of the material that can be modeled remain many orders of magnitude below the duration of a test and the size of a structural test coupon, let alone a structure; therefore, the mechanisms that may be revealed by the model (rather than written into the model explicitly) cannot be guaranteed to be exhaustive of those that arise in large-scale, long-duration experiments. Although bottom-up simulations yield increasingly breathtaking images of small-scale failure mechanisms, they continue to fall well short of representing the complex evolution of damage toward failure in engineering materials.

The top-down method focuses on engineering necessity: Broadly speaking, it seeks always to satisfy the constraint imposed at the structural scale that the displacement fields predicted for given boundary loads match those measured in tests. This can, of course, always be assured for single cases by curve-fitting data; cleverness enters in embellishing the models with representations of the physics of failure that allow prediction for cases (loads and geometry) that have not been tested. The top-down method begins with a macroscopic engineering model, which is progressively augmented by incorporating just those successive levels of detail that are necessary to account for features of engineering tests. Because the model is always calibrated against engineering tests, predictions are available to designers at any stage of model augmentation, subject to some current set of restrictions. Partly for this utility, the top-down method has historically been attractive to engineers. The process of continually validating predictions against tests also rapidly distinguishes those mechanisms or those parameters incorporated in models of mechanisms that have an important effect on engineering performance from those that are irrelevant. This distinction is of practical importance as engineering performance usually depends on only a handful of degrees of freedom, whereas many tens of parameters are commonly proposed in models of mechanisms.

There is much merit in exploring top-down and bottom-up methods concurrently. A profitable strategy is to use the sensitivity analyses of the top-down method to point to those mechanisms for which modeling at finer scales might be pursued, while using the results of bottom-up research to yield the optimal parametric forms to be used when representations of mechanisms are to be incorporated in a top-down model. Regrettably, top-down and bottom-up methods have generally been pursued as distinct exercises by distinct communities, with only occasional serious efforts reported of the transfer of information from one to the other.

In the past decade, research on hierarchical and multiscale models has offered formulations for formally linking top-down and bottom-up approaches in single codes (*13*–*17*). These developments have focused on the mathematical issues of how to embed calculations representing fine-scale phenomena in calculations representing larger-scale phenomena. The mathematical challenges include devising hierarchical meshing strategies that are coarse enough at the largest scales to cover perhaps an entire structure, while cascading down through finer and finer meshes perhaps as far as the atomic scale; integrating classical continuum mechanics calculations with molecular dynamics and even quantum mechanical calculations; and the various boundary-matching problems associated with integrating models executed over different time scales and built on meshes of different gauges.

Hybrid Stress-Strain and Traction-Displacement Models

In continuum (nonatomistic) representations of material damage, two forms of constitutive relation are useful. When spatially continuous

changes arise in the material, including nonlinearity, rate, history, and cycle dependence, the deformation is represented by a constitutive relation between the stress and strain tensors, $\overline{\sigma}\,\overline{\varepsilon}$ (with this abbreviated notation understood to include dependence on time, history, etc.). The law $\overline{\sigma}\,\overline{\varepsilon}$ includes the special case of elasticity and may also include irreversible damage, which appears as hysteresis. The development of relations, $\overline{\sigma}\,\overline{\varepsilon}$, that describe the effects of various kinds of fine-scale damage, including diffuse microcracks, dislocations, and polymer crazing, is the field known as continuum damage mechanics; in this method, discrete damage mechanisms are represented by their effects averaged over suitable gauge lengths, larger than the individual events but small relative to the representative material volume. The second form of constitutive law depicts a discontinuity (or very large gradient) in the material displacement as a localized damage band. A traction-free crack is a familiar special case of a damage band with fully failed material. More generally, the damage band is a mathematically generalized crack across which stresses continue to be transmitted by partially failed material. Although the band in reality may have a finite width, simplification without loss of accuracy (in many cases) can be achieved by representing it as a two-dimensional surface, all the nonlinearity being collapsed into a displacement discontinuity across that surface. As the crack opens, vector tractions, **p**, applied to the crack surfaces equilibrate stresses in the bulk material. The needed constitutive law is the relation **p**(**u**) between **p** and the displacement discontinuity, 2**u**. This relationship is often called a cohesive model.

Materials exhibiting any combination of damage mechanisms can be modeled at the continuum level by hybrid stress-strain and traction-displacement formulations. Furthermore, using a hybrid formulation leads to highly efficient numerical methods. In top-down modeling, the constitutive laws $\overline{\sigma}\,\overline{\varepsilon}$ and **p**(**u**) are treated as functions to be fitted empirically. Eventually, bottom-up models may predict $\overline{\sigma}\,\overline{\varepsilon}$ and **p**(**u**) from first principles; in the near term, bottom-up models provide functional forms for $\overline{\sigma}\,\overline{\varepsilon}$ and **p**(**u**) that guide the empirical fitting process.

Damage Evolution in Structural Composites

As carbon-fiber composites become the first choice for large commercial aircraft structures, the financial motivation for replacing experimental tests by virtual experiments has increased. Both testing labor costs and design cycle times are targets for heavy reduction. But beyond cost, virtual tests also offer the interesting possibility of liberating the design space for composite materials. In the most common strong composite materials, fibers are deployed in unidirectional plies (plies within which all fibers have a common orientation), which are stacked in angled orientations to achieve high stiffness and strength in different directions. Because the stiffness and strength of an individual ply are much higher (by a factor of perhaps 50 to 100) in the fiber direction than in the transverse direction, great performance advantage can be realized by tailoring the ply orientations to suit the anticipated load distribution. In the most general laminate concept, fibers that follow curved paths within plies might be preferred (*18*–*20*), for example, to follow load paths around windows or doors. However, the practicalities of current certification methods force designers to restrict themselves not only to unidirectional plies, but also to just a small number of combinations of ply orientations. The problem is that in an entirely empirical certification system, each variant of ply orientations must be regarded as a distinct material, for which the entire matrix of certification tests must be repeated. If curved fiber paths were used, then in principle an infinite number of different materials would be present in a single structure and certification by empirical methods makes no sense at the material level; certification and the entire range of tests must be done at the level of the entire structure, one candidate fiber pattern at a time!

Thus, structural engineers, limited to ~10^4 tests to certify safety, shy away from complexity, preferring the simplest material solutions that can reasonably do the job (*21*). Simulations that are sufficiently realistic to act as virtual experiments could relax the need for simplicity by vastly increasing the number of tests that can be rapidly and cheaply executed. The essential question, of course, is in how much detail failure mechanisms need to be simulated for a virtual test to give the same engineering outcome as a real test. Minimum requirements are to predict the shape and size of the main crack systems correctly; these dictate the global compliance of the specimen (deflection for given load) and the path to ultimate failure.

The experiments of Figs. 1A and 2A (*22*, *23*) typify laminate material specimen tests in size and ply orientations (0°/90° and 0°/ ±45°/90°). The slot-shaped stress concentrator is often used to represent the kind of severe damage that might be inflicted by a service accident. The circular hole is commonly cut into composite structures to pass bolts, hydraulic and electrical lines, etc. The stress concentrators and potentially the lateral edges of the specimen are sites at or near which damage is likely to initiate. The damage mechanisms shown in Figs. 1A and 2A are some of the most im-

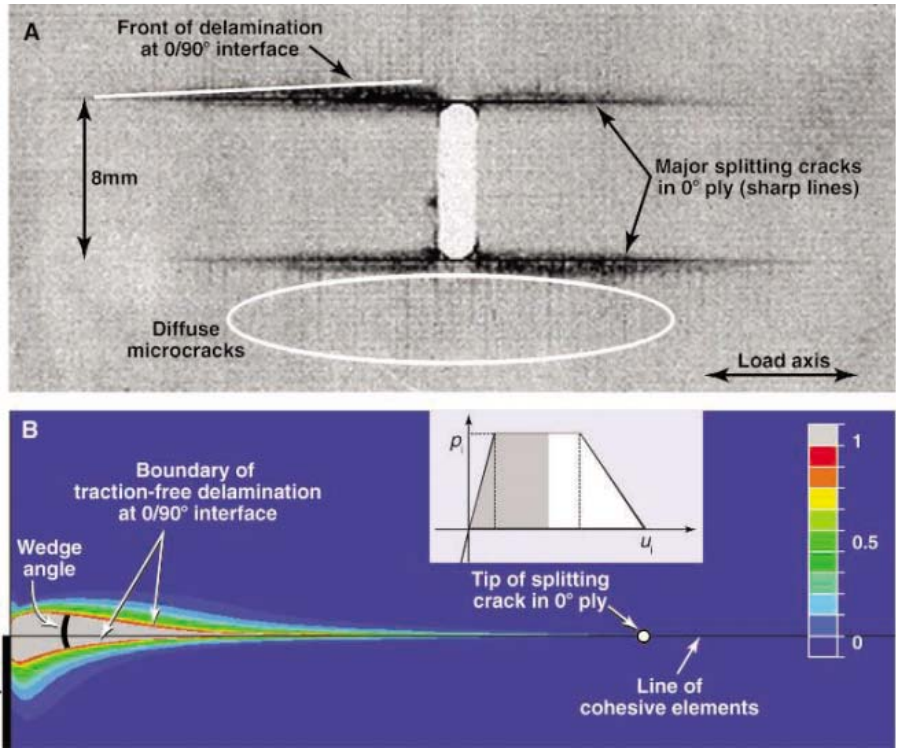

Fig. 1. (**A**) X-ray radiography reveals damage mechanisms viewed through the ply stack in a laminated fibrous polymer composite containing aligned (0°) and transverse (90°) fiber plies loaded in tension (*22*). Dominant splitting cracks appear as sharply defined horizontal lines (in an H configuration) and eventually span the specimen, tending to isolate the strip containing the slit; wedge-shaped delaminations between the plies appear as areas of shadow around the splitting cracks; and myriad microcracks appear between fibers in the transverse plies. (**B**) Plot of level of damage in cohesive model elements between 0° and 90° plies (blue, elastic material; gray, failed material). Inset shows form of cohesive law, for variables defined in text.

portant for tensile loading: splitting cracks, which develop as shear failures; delaminations between plies; and diffuse microcracks running in the fiber direction within individual plies. All of these mechanisms have important effects on the load the composite supports for a given boundary displacement. Splitting and delaminations, together with fiber fractures (which have not yet occurred in Figs. 1A and 2A), also provide paths to ultimate failure. A necessary (minimum) condition for a simulation to be sufficiently realistic is that the evolution of splitting cracks, delaminations, and diffuse microcracking be correctly predicted for arbitrary loads, choices of ply orientation, and shape of the stress concentrator.

Historically, simulations of such notched coupon tests have foundered on some conceptual difficulties with the traditional representations of cracks when they are applied to composites. A very successful model of the elasticity of laminated fibrous composites has been laminate analysis, in which individual plies are homogenized (i.e., the spatial variations in elasticity due to the individual fibers are averaged out) but the heterogeneity from ply to ply is retained. However, in the presence of a free edge, such as at the periphery of a cutout, the ply heterogeneity implies that a singularity must exist in the elastic fields at ply interfaces (*24*). The singular fields (strain energy concentrations) suggest correctly that the interfaces will be the sites of cracking, but the question of how to model the first damage and its progression into a delamination crack of detectable size has proven very thorny.

In linear elastic fracture mechanics (LEFM), which is the traditional model of cracking in nonductile materials, an idealized traction-free crack is assumed and all the material toughness that resists crack advance (the fracture toughness) is assigned to a point process located at the crack tip (Fig. 3); the rest of the body is assumed to be linear-elastic. The beauty of LEFM is that for cracks that are relatively large, the fracture toughness proves to be a material constant, and LEFM, once calibrated, can predict the external loads at which the crack tip conditions will cause crack propagation for any component geometry. LEFM has been remarkably successful in engineering design, including the design of composites, provided a sufficiently large crack is already present. Conservative and therefore safe design can be achieved by assuming that a crack big enough to be detected in service inspections is present and requiring that the load that LEFM predicts for crack propagation exceeds service loads. However, creating realistic damage simulations requires much more than this. The initiation of damage at stress-concentrating sites must be predicted for parts that contain no cracks, and the gradual evolution of initiated damage into large cracks must be modeled.

The critical conceptual limitation of LEFM is representing all the material nonlinearity during crack extension as a point process. In fact, the zone of nonlinearity always has finite size. Nonlinear fracture models, and in particular the cohesive zone idealization, describe the development of the zone of nonlinearity explicitly (Fig. 3) (*25*–*29*).

Modeling Multiple Mechanisms

Various modeling attempts in the recent literature (*30*–*37*) have led to an initial list of necessary model features for simulating notched laminate failure under tensile loads, judged according to the top-down philosophy by whether the attempt has succeeded or failed to reproduce engineering tests. A summary of necessary features can be illustrated through simulations of the tests of Figs. 1A and 2A (*37*). The simulations are based on the finite element method, which computes stress distributions for generic geometry and loads. Special model features are embedded within the finite element formulation by the use of custom elements and nonlinear constitutive laws.

In these simulations, delamination damage is modeled by a cohesive model with a reasonable functional form for $\mathbf{p}(\mathbf{u})$ [Fig. 1B, inset; $u_1 \geq 0$; $p_i(u_i) = -p_i(-u_i)$, $i = 2, 3$, where p_i and u_i are components of $\mathbf{p}$ and $\mathbf{u}$ and the Cartesian coordinates 2 and 3 lie in the plane of the crack] independently determined by the analysis of polymer adhesives in de-adhesion experiments (*38*). The cohesive model is incorporated in the simulations by special interface elements, which allow a displacement discontinuity when the local stress satisfies a critical condition. The elements are planted over all ply interfaces to allow the possibility of ply delamination with nonprescribed delamination shape. In the first simulations to be described, they are also planted along the planes within the 0° plies that are tangent to the ends of the slot, where the dominant splitting cracks occur in Fig. 1A. A key feature of the cohesive model is that it allows arbitrary mixed-mode crack tip conditions—that is, arbitrary proportions of opening and sliding displacements across the nonlinear process zone—which can (and do) vary along a crack front. Calculating the separation of the energy released by crack advance into contributions attributable to crack opening and sliding, which is prerequisite to predicting the condition for crack

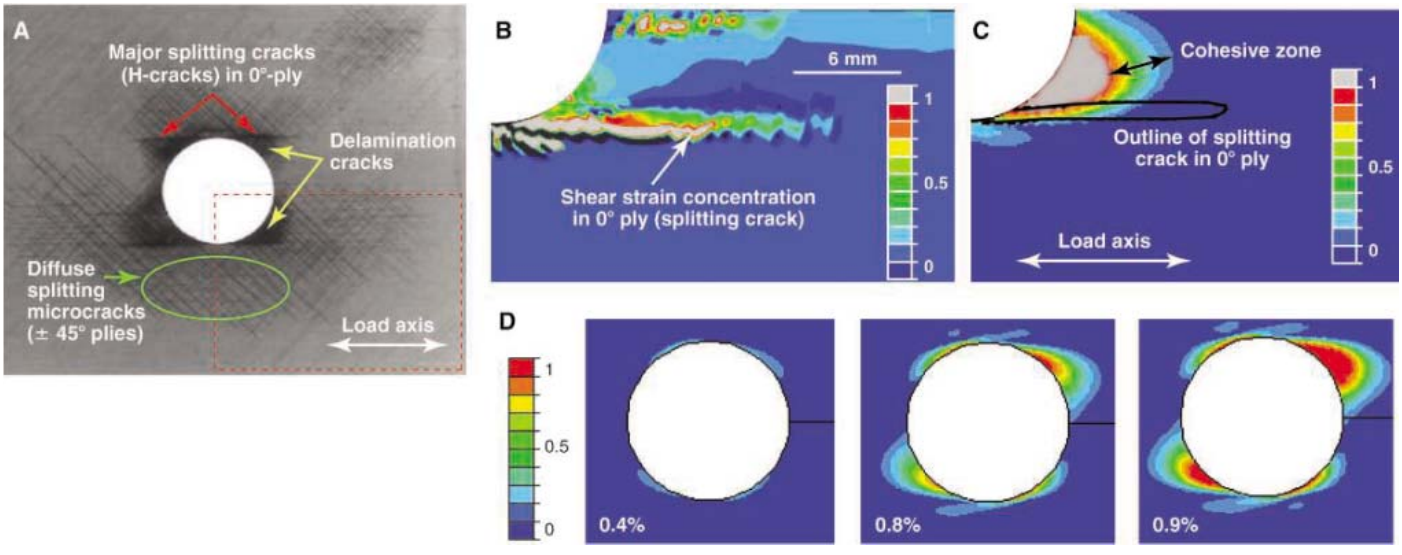

Fig. 2. (**A**) Images similar to Fig. 1 for a laminate that also contains angled (±45°) fiber plies and a circular hole instead of a slit (*23*). The splitting cracks are now shorter, the delaminations are no longer wedge-shaped but lobe-shaped, and diffuse microcracking occurs predominantly in the ±45° plies. (**B**) Computed continuum damage distribution in 0° ply (blue undamaged; gray is completely failed material) showing shear strain concentration that forms splitting crack. (**C**) Computed damage in cohesive zone between 0° and 45° plies (blue is undamaged; gray is completely failed material). (**D**) Initiation and evolution of damage in cohesive zone between 0° and 45° plies at marked applied strains. Blue is undamaged material; red is completely failed (traction-free).

propagation, is very troublesome for cracks of general shape in the LEFM formulation, implemented for example via the virtual crack closure technique. In a cohesive model formulation, the energy partition is immediately deducible from the opening and sliding displacements, which are computed automatically as part of the finite element problem. A suitable failure criterion built into the cohesive model then allows automatic calculation of the evolution of crack shape (*37*).

Diffuse microcracking, which may occur in any ply, is modeled by assigning a nonlinear constitutive law, $\overline{\sigma}\,\overline{\varepsilon}$, to the ply material that causes material degradation (softening) according to a continuum damage model. Many models of reasonable functional form are available to represent this effect [e.g., (*39*, *40*)].

Even though the specimens of Figs. 1 and 2 are thin sheets loaded in-plane, opening displacements arise when cracking occurs. Therefore, accurate calculation of the crack displacements requires that the full three-dimensional stress state be calculated. Plate elements, which have been very popular in laminate analysis because they greatly reduce the number of degrees of freedom in a calculation, are not adequate; three-dimensional finite elements should be used. For mesh-independent results, these elements must not be larger than the nonlinear process (cohesive) zones that arise at crack tips during a calculation, which in a typical aerospace carbon-epoxy laminate are ~0.3 to 1 mm. Fortunately, the zone lengths can be estimated in advance from analytical cohesive model results (*27*, *37*, *41*), avoiding the need for iterative mesh refinement. If integrations are performed with carefully developed numerical methods, especially where cohesive zones extend partway across an element, then it is sufficient for the three-dimensional elements used for plies to be commensurate with (rather than smaller than) the cohesive zone lengths (*37*).

Simulations that follow all these rules (a qualification that excludes most of the literature) are encouragingly successful. The wedge-like shape of the delamination cracks in the 0°/90° laminate is reproduced, with comparable wedge angle (allowing uncertainty about how far the dye penetrant used in the x-ray measurements infiltrated the damage zone), and the length of the splitting cracks is in approximately the correct proportion to the delamination size. Other plots show that the distribution of diffuse microcracking in the 90° plies is predicted correctly within the uncertainty of the image resolution.

The importance of mutual interactions among different failure mechanisms due to the effect of each on the stress field experienced by the others can quickly be shown by turning off one mechanism in a simulation (*36*, *37*). Thus, for example, when ply nonlinearity is omitted from the calculation, the delaminations of Fig. 1A are predicted incorrectly to grow mainly between the dominant splitting cracks instead of outside them (*37*). When ply nonlinearity is included, the zone in the 90° ply in which it is concentrated coincides with the area of the delamination between the 0° and 90° plies at any instant.

The instructive possibility of nonuniqueness in formulating a model that is sufficient from the top-down view is highlighted in the simulations of the test of Fig. 2A. In these simulations, no cohesive elements were planted along the crack paths that are expected to be followed by splitting cracks. But a manifestation of the splitting cracks still arises: A shear damage band has formed in the same locations (Fig. 2B), mediated by the continuum deformation introduced by the relation $\overline{\sigma}\,\overline{\varepsilon}$ assigned to the 0° plies. Provided $\overline{\sigma}\,\overline{\varepsilon}$ includes the possibility of material failure, the damage band is not only highly localized (because of instability in the local stress state caused by the stress-concentrating hole) but also can evolve into a traction-free crack. From the point of view of the fine-scale material behavior, a discrete crack model and a continuum damage representation are very different. But the outcome at the level of the engineering performance of the structure in this particular case is much the same.

The level of fidelity shown in Figs. 1 and 2 is a recent accomplishment (*37*). When the ply orientations and hole shape are changed, the same model used in Fig. 1, without any parameter modification other than the choice of representing splitting cracks, correctly predicts the new damage patterns (Fig. 2, B and C), especially the lobe shape of the delaminations and their size in proportion to that of the splitting cracks. In contrast, prior model formulations failed to reproduce crack shapes correctly, usually because the mesh choice did not satisfy the requirement of not being larger than the damage zones, so that the ratio of sliding to opening displacement across the zone could not be predicted, or the three-dimensional stress state was not fully modeled, or the interactions between all three mechanisms present in the experiments were not considered.

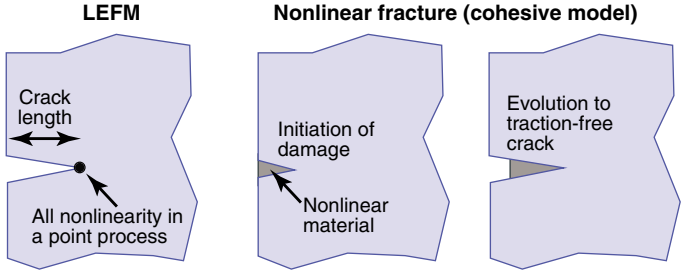

Fig. 3. According to linear elastic fracture mechanics (LEFM), the applied load required to evolve damage (grow the crack) tends to infinity as the crack length approaches zero. In a cohesive model, the applied load for damage initiation and evolution is limited to some finite multiple of the maximum value p_{max} of a component of the assigned cohesive law **p**(**u**).

The cohesive model of delamination provides a unifying physical model of damage initiation and its progression to large cracks (Fig. 2D) and is an important conceptual advance in damage modeling. Initiation sites are determined by local stress conditions; the delamination damage zones then extend both around and away from the stress concentrator, until the displacement discontinuity vector exceeds the critical condition for forming a traction-free zone. When the traction-free zone becomes large enough, the limit in which LEFM is valid is approached and the two models become equivalent.

The simulations of Figs. 2 and 3 are only part of the whole story: Given the state of damage predicted for the initial tensile load, one can then restate the boundary conditions to ask how damage will evolve for other subsequently applied load types, including compression and twist. The application of compression after loads that have caused delamination is particularly dangerous: Local ply buckling causes further delamination growth and is a primary failure mode for damaged airframes and marine structures. The critical compressive load for ply buckling is approximately inversely proportional to the delamination size and depends on the delamination shape.

The Extended Finite Element Method: Dynamic Crack Path Selection

For splitting cracks originating at notch extremities and delamination cracks, the potential crack paths are known a priori because they are constrained by the geometry of either the material or the specimen. Other crack systems, including splitting cracks in specimens of more complicated geometry, shear bands, and distributed intraply matrix cracks, follow paths that are determined during damage evolution by changes in the local stress fields. An important advance in finite element formulations, the extended finite element method (XFEM), defines solid continuum elements within which a general displacement discontinuity can be introduced if some failure criterion is satisfied in the element at any time during a simulation (*42*–*44*). The displacement discontinuity can appear on any plane in the element and can be associated with tractions, **p**. Thus, cracks described by a cohesive crack model can appear in any number and follow any path during a simulation, determined by the evolving stress distribution. Crack branching and mother-daughter crack systems can appear. Implementation of three-dimensional elements in the XFEM formulation is a current research activity and will soon add an important empowerment to damage simulations.

Calibration and Validation

Even as the successful prediction of delamination crack shapes shown in Figs. 1 and 2 encourages the quest for virtual experiments, designers require

simple methods of calibrating and validating the nonlinear constitutive laws, $\mathbf{p}(\mathbf{u})$ and $\bar{\sigma}\bar{\varepsilon}$. The cohesive law is especially challenging, because it refers for polymer composites to nonlinear zones that are ~1 mm or less in size. The inference of $\mathbf{p}(\mathbf{u})$ from the nonlinearity in load-deflection data, which works well for materials with larger cohesive zones of ~10 mm (*45*, *46*), is problematic for a 1-mm zone because specimen deflections are small during the zone's development. Once the crack is relatively large (well-developed damage zone), its behavior contains no information about $\mathbf{p}(\mathbf{u})$ other than $\int p_i du_i$ (the work of fracture) (*27*, *47*). Alternative experiments that infer $\mathbf{p}(\mathbf{u})$ from crack profiles in plane specimens such as the short shear beam or cantilever beam (*46*, *48*) would need to resolve displacements around cracks that are barely discernible. Thus, none of the common methods in the literature appear immediately workable. New experimental methods for measuring very small crack displacements (e.g., with high-resolution x-ray tomography) are very promising; however, the interpretation of such experiments may prove challenging, which raises the question of whether x-ray tomography can serve as a standard calibration method executed by a field engineer. A simpler prospect may be to use the evolution of the macroscopic crack shape as the calibrating information, because Figs. 1 and 2 suggest that this is an information-rich experiment for determining $\mathbf{p}(\mathbf{u})$. Current research continues to seek a calibration method, validated by testing the accuracy to which ensuing predictions match fracture experiments.

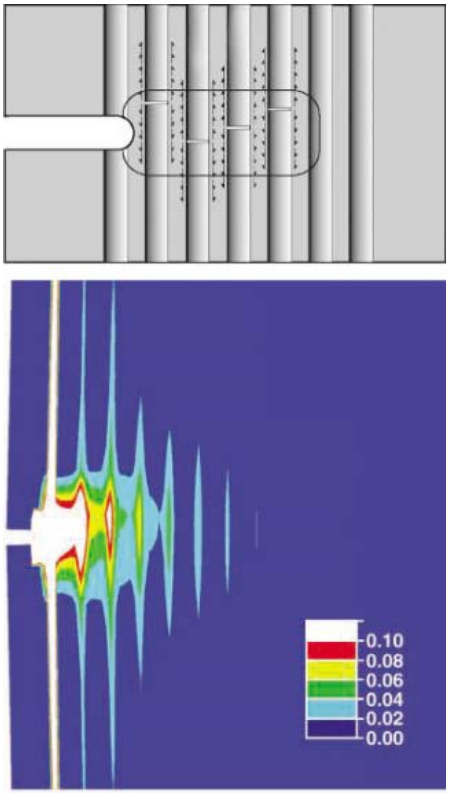

Fig. 4. Simulation of a metal matrix composite failing in bending: stochastic fiber breaks and interfacial sliding (**top**) and accumulated plastic strain (second strain invariant—scale given in key) in the matrix (**bottom**) (*49*).

Fiber Bundle Failure

The tests of Figs. 1 and 2 will ultimately terminate in fiber bundle failure. Simulating fiber failure requires further model augmentation, with a locally refined mesh in which fibers are represented discretely. Constitutive laws must be added at the fiber scale to represent fiber failure, debonding of fibers from the matrix and frictional sliding between the two, and matrix deformation and failure. Thus, a hybrid stress-strain and traction-displacement formulation is again appropriate. In the recent example reproduced in Fig. 4 (*49*) for a metal matrix composite of relatively large SiC fibers in a Ti-6Al-4V alloy matrix, one cohesive model was used to control fiber failure and a second to model fiber-matrix interfacial friction, while continuum laws modeled matrix deformation. (The friction law includes resistance to material interpenetration in the relation between $\mathbf{p}$ and $\mathbf{u}$; sliding resistance is described by a relation between the normal and shear components of $\mathbf{p}$.) The simulation reveals details of the sequence of fiber failure events and the severe matrix deformation that is caused by consequent load shedding. Using material properties known a priori, load-displacement data for a notched bend test could be reproduced, including nonlinearity, ultimate strength, and snap-back phenomena associated with the rupture of individual fibers.

Current Challenges

The ability to link separate resin and fiber properties to the engineering metrics of fiber bundle rupture raises the attractive prospect of predicting the effect of material choice via a virtual experiment. However, this is more complicated than the idealization of Fig. 4 suggests. First, the reality of fibrous reinforcement is that fibers appear in random rather than spatially ordered patterns, with substantial variations in the nearest-neighbor spacing that are not easily quantified. Although this may have a minor effect on fiber rupture under aligned loads, it can dominate the cracking sequence and the statistics of crack initiation under transverse loads (*50*). Second, finer fibers than the SiC fibers of Fig. 4, such as 7-μm carbon fibers, follow paths that are slightly wavy rather than ideally straight. The statistics of fibers that are misaligned over a critical volume exceeding ~1 mm (*51*) control compressive failure via the mechanism of fiber kinking (fiber buckling instability due to fiber rotation) (*52*–*54*). Third, fine fibers tend to mingle from one nominally unidirectional ply to another, forming mechanical bridges across delamination planes, with important consequences for the cohesive tractions that control the delamination. The state of the art of top-down models is that all such stochastic details of microstructure are subsumed in the constitutive laws used in coarser-scale models. To incorporate fiber-level mechanisms in fiber-scale models, substantial research is still required to relate distributions in geometrical and material parameters to the statistics of test data.

One can also contemplate interfacial debonding and the complexity of static and dynamic friction, as well as the molecular-level mechanisms (such as polymer microcracking and crazing) that are the fundamental mechanisms of mixed mode delamination. With the formalism of multiscale methods available, these are the next challenges for creators of virtual experiments. But observe the propensity of top-downers to fit engineering data with a large number of parameters and claim that the resulting model is unique and imbued with predictive power, and that of bottom-uppers to project that their favorite mechanism is the one that propagates up among all others to control engineering behavior. The challenges will not be easily met; beware premature claims of victory.

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REVIEW

Nanoparticle Polymer Composites: Where Two Small Worlds Meet

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The mixing of polymers and nanoparticles is opening pathways for engineering flexible composites that exhibit advantageous electrical, optical, or mechanical properties. Recent advances reveal routes to exploit both enthalpic and entropic interactions so as to direct the spatial distribution of nanoparticles and thereby control the macroscopic performance of the material. For example, by tailoring the particle coating and size, researchers have created self-healing materials for improved sustainability and self-corralling rods for photovoltaic applications. A challenge for future studies is to create hierarchically structured composites in which each sublayer contributes a distinct function to yield a mechanically integrated, multifunctional material.

The mixing of nanoparticles with polymers to form composite materials has been practiced for decades. For example, the clay-reinforced resin known as Bakelite was introduced in the early 1900's as one of the first mass-produced polymer-nanoparticle composites (*1*) and fundamentally transformed the nature of practical household materials. Even before Bakelite, nanocomposites were finding applications in the form of nanoparticle-toughened automobile tires prepared by blending carbon black, zinc oxide, and/or magnesium sulfate particles with vulcanized rubber (*2*). Despite these early successes, the broad scientific community was not galvanized by nanocomposites until the early 1990s, when reports by Toyota researchers revealed that adding mica to nylon produced a five-fold increase in the yield and tensile strength of the material (*3*, *4*). Subsequent developments have further contributed to the surging interest in polymer-nanoparticle composites. In particular, the growing availability of nanoparticles of precise size and shape, such as fullerenes, carbon nanotubes, inorganic nanoparticles, dendrimers, and bionanoparticles, and the development of instrumentation to probe small length scales, such as scanning force, laser scanning fluorescence, and electron microscopes, have spurred research aimed at probing the influence of particle size and shape on the properties of nanoparticle-polymer composites.

As part of this renewed interest in nanocomposites, researchers also began seeking design rules that would allow them to engineer materials that combine the desirable properties of nanoparticles and polymers. The ensuing research revealed a number of key challenges in producing nanocomposites that exhibit a desired behavior. The greatest stumbling block to the large-scale production and commercialization of nanocomposites is the dearth of cost-effective methods for controlling the dispersion of the nanoparticles in polymeric hosts. The nanoscale particles typically aggregate, which negates any benefits associated with the nanoscopic dimension. There is a critical need for establishing processing techniques that are effective on the nanoscale yet are applicable to macroscopic processing. Another hurdle to the broader use of nanocomposites is the absence of structure-property relationships. Because increased research activity in this area has only spanned the past decade, there are limited property databases for these materials (*5*). Thus, greater efforts are needed to correlate the morphology of the mixtures with the macroscopic performance of the materials. Establishing these relationships requires a better understanding of how cooperative interactions between flexible chains and nanoscopic solids can lead to unexpected behavior, like the improved mechanical behavior of mica-reinforced nylon.

The interactions of nanoparticles with polymers are mediated by the ligands attached to the nanoparticles; thus, the ligands markedly influence particle behavior and spatial distribution. Therefore, we first describe recent advances in nanoparticle surface modification and then the interactions between these particles and polymer matrices, including homopolymers, diblock copolymers, and blends.

Surface Functionalization of Nanoparticles

The surface chemistry of nanoparticle functionalization evolved in part from studies on functionalized planar surfaces, including self-assembled monolayers (SAMs) (*6*) and polymer brushes (*7*) on substrates ranging from gold to metal oxides. As with planar substrates, functional small molecules and polymers can be attached to nanoparticles by physical adsorption or covalent attachment. Synthetic strategies that give polymer-functionalized nanoparticles include performing the particle synthesis directly in the polymer matrix, replacing small-molecule ligands inherent to a nanoparticle synthesis with functional polymers in a "grafting-to" process, and growth of polymers from functionalized nanoparticles in a "grafting-from" process. It is imperative, though, that the conditions used retain the specific characteristics of the nanoparticles.

Many state-of-the-art nanoparticle preparations afford high-quality particles by decomposition of organometallic precursors in a solution environment that leads to surface coverage with small-molecule alkane-based ligands, like *n*-alkyl thiols, amines, or

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phosphine oxides. Simply mixing these alkane-covered nanoparticles with most polymers leads to nonuniform particle clustering or aggregation that compromises the properties of the composite. Replacement of these conventional ligands with those that favorably interact with the polymer host is key in achieving uniform dispersions. Thus, versatile synthetic strategies are necessary to fine-tune the chemical nature of the ligands. Grafting-from polymerization requires replacing the alkane ligands with functionality that supports polymerization radially outward from the nanoparticle surface. Mild polymerization techniques, such as ring-opening metathesis polymerization (ROMP) and controlled free-radical polymerizations, have proven very useful in grafting-from experiments, giving well-dispersed nanoparticles within the composite (*8*, *9*). On the other hand, the grafting-to approach can be simpler to perform and is useful with low-molecular-weight polymer ligands or when the nanoparticles are sensitive to polymerization conditions (*10*). Grafting-to chemistry has been particularly effective for preparing polyethylene glycol-grafted (PEGylated) nanoparticles, which imparts hydrophilicity and biocompatibility to the nanoparticles (*11*–*14*).

Nanoparticles and Homopolymers

The simplest of filled polymers, nanoparticles in a single-component melt, are particularly valuable for obtaining insight into the fundamental factors that control particle dispersion. Although enthalpic interactions are usually dominant (*15*, *16*), entropic interactions can play a critical role in dictating structure. Computational studies on filled homopolymers near a surface containing a notch or crack showed that the polymer melt induced an entropic "depletion attraction" between the particles and surface that drives a fraction of the nanoparticles into the defect (*17, 18*). These predictions were recently demonstrated by experiments on a multilayer comprised of PEGylated CdSe nanoparticles dispersed in poly(methyl methacrylate) (PMMA) in contact with a brittle silicon oxide layer (*19*). Heating the bilayer above the glass transition temperature of the composite induced cracking of the SiO_x layer, and nanoparticles that were comparable in size to the radius of gyration of the PMMA migrated into the crack, as shown in the fluorescence optical micrograph in Fig. 1. Notably, for smaller nanoparticles, this behavior was not observed, because the polymer can easily accommodate the nanoparticles without a substantial entropic penalty. These findings open a convenient route for designing self-healing systems.

Similar entropic interactions were operative in polystyrene (PS)–functionalized CdSe dispersed in PS examined by Crosby and co-workers (*20*), in which nanoparticles were found to segregate to the tip of a crack (Fig. 2) and modify the crazing characteristics of a glassy polymer. Nearly a doubling of the strain-to-failure was found, but a quantitative understanding of the origins of the enhanced properties is still lacking. Nanoparticles, in particular the platelike nanoparticles or nanosheets of clay, have been shown to enhance markedly the mechanical properties and reduce permeability of polymers that crystallize (*3*, *4*).

Composites based on conductive or semiconductive nanoparticles are very attractive for electronic applications and can augment the use of conjugated organic polymers in flexible electronics, light-emitting displays, and photovoltaics. However, conventional alkane ligands insulate nanoparticles from their surroundings, which greatly diminishes their performance. Organic-inorganic hybrid photovoltaics composed of polythiophene–quantum dot (or CdSe nanorod) composites show enhanced performance with tailored ligand coverage, such as amine- and phosphonic acid–terminated polythiophenes (*21*). The enhanced dispersion of particles or nanorods in the polythiophene matrix improves device performance as manifest in significantly improved power conversion efficiencies that are ~3 times that of conventional blends but use only half the nanocrystalline material by volume.

Although uniform nanoparticle dispersions are desired for many applications, particle aggregation can also be used to advantage. Consider CdSe nanorods functionalized with alkane ligands in PS. Because of the incompatibility of the ligands with the PS, the system phase separates into domains of PS and nanorods. If an electric field is applied to a solution of nanorods mixed with PS, the nanorods align in the direction of the applied field and, as solvent evaporates, the PS and oriented nanorods phase separate. Minimization of the interfacial energy between the domains drives a close packing or "self-corralling" of the nanorods into a hexagonal array oriented normal to the film surface (*22*, *23*) (Fig. 3). Such assemblies are currently being investigated for use in photovoltaic devices where self-corralling is used in concert with surface patterning and interfacial interactions to direct the lateral positioning of the ordered arrays of nanorods onto an underlying circuit.

Nanoparticles have also been used to modify the flow characteristics of homopolymer melts. Mackay and co-workers (*24*) demonstrated that the addition of small amounts of nanoparticles to homopolymers can eliminate "shark-skin" effects, instabilities generated during the extrusion of polymers. In thin polymer films, large nanoparticles can be used to nucleate dewetting, whereas smaller nanoparticles have been found to retard dewetting (*25, 26*). In each of these cases, the segregation of the nanoparticles to the interface between the polymer and the solid surface alters the flow conditions at the interface.

Nanoparticles and Block Copolymers

Diblock copolymer/nanoparticle mixtures have attracted substantial attention because the microphase separation of the copolymer can direct the spatial distribution of nanoparticles and thereby tailor the properties of the composite. The optical performance of composites, for example, is highly sensitive to the specific location of the particles within the matrix (*27*), and the microphase separation of block copolymers can be used to great advantage. However, block copolymers do not simply "template" the arrangement of the nanoparticles (*28*). The nanoparticles can act on the block copolymer, and nanoparticles have been found to alter both the orientation (*29*, *30*) and the morphology (*31*–*34*) of the diblock copolymer microdomains. Theoretical and computational modeling efforts revealed two important features of these composites: (i) the role that excluded volume effects of relatively large nanoparticles play in the structure of the composite and (ii) the effect of the particle dispersion on the macroscopic properties of the composite.

Because the nanoparticles are solids, the polymer chains must stretch around these obstacles, causing a loss in conformational entropy that increases with particle radius (*35*). In the absence of specific interactions, larger nanoparticles are expelled from the bulk of the copolymers, whereas smaller particles are not. This significantly affects the spatial distribution of nanoparticles within homopolymers and block copolymers and the global structure of the particle-filled systems. Using

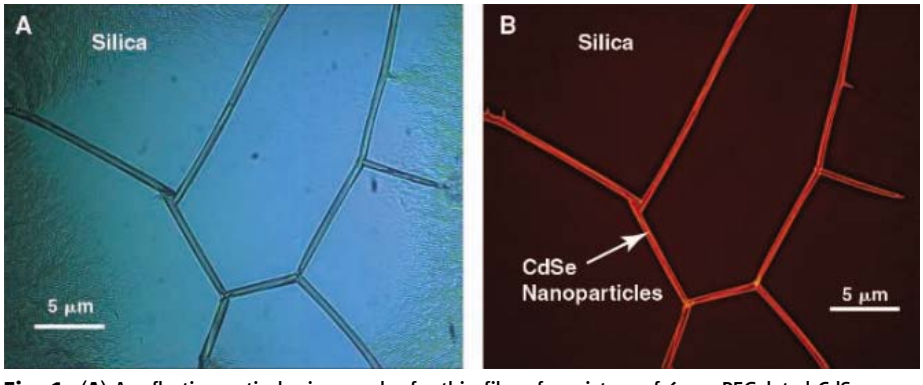

Fig. 1. (**A**) A reflection optical micrograph of a thin film of a mixture of 6-nm PEGylated CdSe nanoparticles in poly(methylmethacrylate) that was spin-coated onto a silicon wafer. A 0.1-μm layer of silicon oxide was evaporated onto the nanoparticle composite and the trilayer was heated to 160°C for 4 hours. The difference in the thermal expansion coefficients of silicon oxide and the PMMA composite produced the cracks observed. (**B**) The same film viewed with a fluorescence microscope, in which the segregation of the CdSe nanoparticles to the cracks is highlighted by the fluorescence of the nanoparticle.

a combination of self-consistent field- and density-functional theories, Thompson *et al*. predicted that larger A-like particles (i.e., particles that are compatible with the A blocks of AB copolymers) localize at the center of the A microdomains, whereas smaller particles are more uniformly dispersed within a specific microdomain (*35*). O'Shaughnessey *et al*. arrived at similar conclusions, using analytical scaling theory to investigate the behavior of particles in grafted layers (*36*).

The observations above imply that the spatial distribution of nanoparticles in the microphase-separated morphologies can be controlled by tailoring the nanoparticle ligands (i.e., enthalpic effects) and the size of the nanoparticles relative to the radius of gyration of the polymer (i.e., entropic effects). For example, by blending A-like nanoparticles of different sizes with symmetric AB diblocks, one can fashion gradient materials within the A lamellae, where the largest particles are localized in the center, bordered by the next larger particles, which in turn are neighbored by smaller particles (*37*). Such gradient materials result in unusual optical or electrical properties (*27, 38*).

The entropic penalty associated with chain stretching around larger particles can also give rise to transitions from one microphase to another. For example, Lee *et al*. used theory and computational modeling to show that, at fixed diblock composition, interaction energies, and particle volume fractions, an increase in particle size is sufficient to force a lamellar morphology into a cylindrical morphology (*31*). Recent experimental studies have confirmed these predictions (*32–34*).

When particle-filled diblock films are confined between two surfaces, the effects of chain stretching again play an important role. Using a combination of theoretical approaches, Lee *et al*. considered a symmetric AB diblock copolymer confined between two A-like surfaces (*29*). In the absence of nanoparticles, when the lamellar period of a symmetric diblock copolymer is commensurate with the separation distance between the confining walls, the microdomains orient parallel to the interfaces. The addition of nanoparticles, however, causes the microdomains to reorient normal to the walls. Recent experimental studies showed exactly this behavior where nanoparticles were expelled to the surfaces, mediated interfacial interactions, and caused the microdomains to orient normal to the surface (*30*).

By combining such entropic effects with enthalpic interactions, it is possible to achieve even greater control over the morphology of the composites. For example, if the system contains particles that differ not only in size but also in surface chemistry, one can carefully tailor nanoparticle placement within a diblock matrix (*39*, *40*) and thereby form structures that are decorated with particles in a well-defined manner. Enthalpic interactions between the blocks and functionalized nanoparticle surfaces can be exploited to achieve precise nanoparticle placement. Kramer and coworkers (*41*, *42*) used thiol-terminated polymers on gold nanoparticles to create "neutral" or nonselective particles that localized at the interface between the PS and poly(2-vinylpyridine) (P2VP) microdomains, (Fig. 4) reducing interfacial energies and inducing a transition from a lamellar to a bicontinuous morphology. These findings are in agreement with recent computational studies indicating that nanoparticles can stabilize bicontinuous phases in simple binary mixtures (*43*).

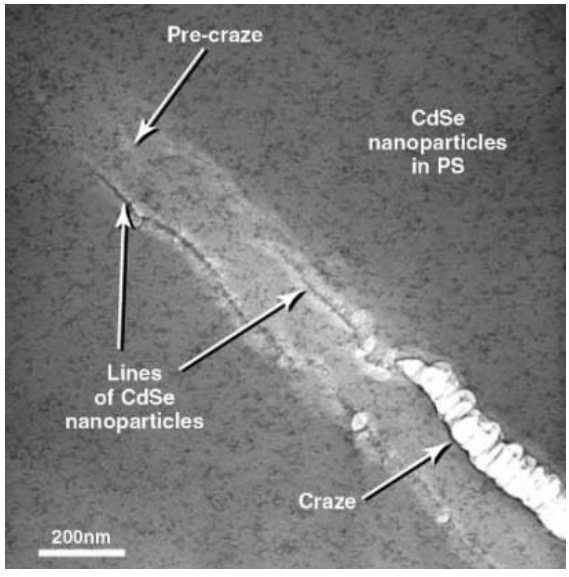

Fig. 2. Transmission electron microscopy (TEM) image of 5-nm CdSe/ZnS nanoparticles with polystyrene (PS) ligands in polystyrene where a craze has been introduced to the film. A uniform dispersion of the nanoparticles in the PS matrix is seen outside the crazed region, but in the precrazed zone, the nanoparticles are seen to line up in front of the craze. [After (*20*), courtesy A. Crosby]

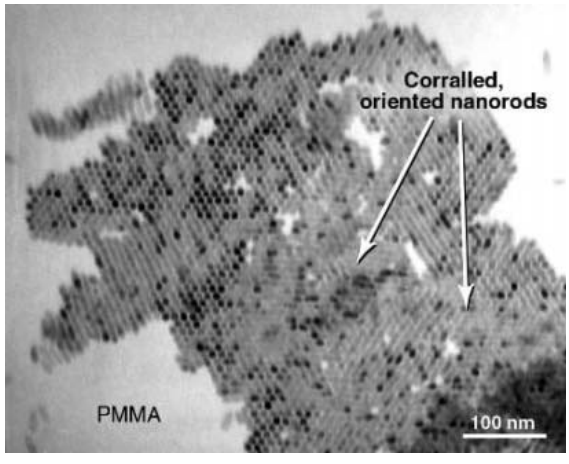

Fig. 3. A TEM image of an ordered hexagonal array of 8-nm diameter CdSe nanorods (40 nm in length) that resulted when a mixture of the nanorods with alkane ligands was mixed with PMMA in chloroform. The solvent was evaporated under an applied electric field of 10^7 V/m. The electric field aligned the nanorods, whereas the phase separation for the nanorods with PMMA resulted in a corralling of the nanorods as a result of the interfacial tension between the nanorod phase and the PMMA.

Recent computational studies have examined the role of the spatial distribution of nanoparticles within a material on its optical (*44*) and mechanical properties (*45*, *46*). Buxton *et al*. simulated the propagation of light through a diblock/nanoparticle composite (*44*) to show that the optical properties are highly dependent on the spatial distribution of the fillers within the polymer matrix and provided guidelines for tailoring the particles and polymers to yield the desired performance. Thompson *et al*. used a self-consistent field approach to predict the effect of nanoparticle additives on the elastic properties of a block copolymer melt (*45*). Further measurements, though, are needed to correlate the morphology of the particle-filled diblocks to their properties.

Nanoparticles and Mixtures

Recent investigations of nanoparticles in immiscible mixtures have focused on using the particles to stabilize evolving morphologies and/or arrest domain coarsening. For example, Lin *et al*. demonstrated that nanoparticles can behave like surfactants, localizing at the interfaces in oil/water mixtures (*47*). More specifically, the particles formed a monolayer around the dispersed droplets, allowing the fabrication of novel nanoparticle capsules. Such interfacial activity can also be exploited in polymer blends. Rafailovich and co-workers (*48*) observed a significant reduction in the domain size in phase-separating binary blends using clays modified with organic ligands as a result of the localization of the clay sheets at the interface between the immiscible components. Controlling the domain size in this manner improves the overall mechanical integrity of the material.

Nanoparticles have also been shown to influence the phase-separation kinetics in polymer mixtures. Tanaka *et al*. showed that nanoparticles can significantly alter the coarsening dynamics of mixtures (*49*). Theoretical studies of nanoparticle-filled mixtures suggest the existence of distinct pattern formation at early stages of phase separation (*50*, *51*) and a subsequent slowing of domain growth at later times (*52*). Experiments of Composto *et al*. (*53*) and Krishnamoorti *et al*. (*54*) confirmed a substantial slowing of phase separation with the addition of nanoparticles to a polymer mixture.

Tanaka *et al*. (*55*) recently used computer simulations to reveal unexpected morphological transitions in symmetric AB blends with the addition of

mobile nanoparticles. At high particle concentrations, insufficient amounts of A to cover all the A-like particles cause a "wetting-induced depletion" that gives rise to unusual reentrant morphological changes as a function of particle concentration. So the volume fraction provides a simple means to control the morphology of particle-filled, immiscible mixtures.

Computer simulations have also been used to investigate the influence of nanorods on the evolution of phase-separating AB binary blends (*56*). For rods coated with A-like chains that are immersed in an AB mixture with a minority A phase, a continuous network evolves at very low nanorod volume fractions. The rods form a percolating network within A, with the percolation threshold being essentially half the value found in a single component fluid.

There have been relatively few systematic experimental or theoretical studies of the thermodynamic properties of mixtures of nanoparticles and binary polymer blends. Consequently, there is little quantification of the influence of nanoparticle size, volume fraction, or chemical nature on the phase behavior of the system. Ginzburg (*57*) and He *et al.* (*58*) recently carried out thermodynamic calculations to determine the influence of nanoparticles on the miscibility of blends of A and B homopolymers. By decreasing the size of the nanoparticles, a phase-separating system can be driven into a thermodynamically miscible, one-phase region. Thus, by tailoring the particle size, desired phase behavior can be controlled (*58*).

As in the case of nanoparticle-filled diblocks, a critical challenge lies in predicting how the morphology of the filled blends affects the macroscopic properties of the composite. To date, there have only been a few theoretical studies in this area. Buxton *et al.* (*59*) integrated different computational approaches to relate the morphology of a particle-filled mixture to mechanical behavior and electrical conductance. In these rod-filled binary blends, the reinforcement efficiency of the nanorods and the electrical conductivity of the materials were significantly higher than the respective behavior for homopolymer composites.

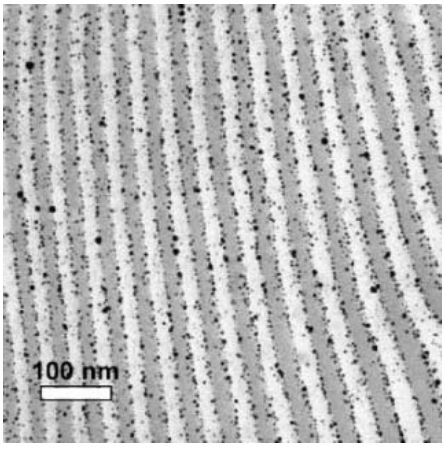

Fig. 4. Cross-sectional TEM image of a symmetric polystyrene-b-poly(2-vinylpyridine) diblock copolymer containing gold nanoparticles with PS ligands with a nanoparticle volume fraction of 0.28. The segregation of the nanoparticles to the interface between the microdomains of the copolymers is immediately evident in the micrograph. [Used with permission (*20*)]

Conclusions

Central to discussions of nanoparticle-polymer composites is a consideration of enthalpic and entropic interactions when functionalized nanoparticles are introduced into polymers. Examples were highlighted in which entropic interactions were exploited to direct the placement of nanoparticles, tailor morphologies, and thereby control the macroscopic behavior of the composite material. Through surface modification of nanoparticles, it is also possible to vary the enthalpic interactions in the system such that structural organization within nanocomposites can be achieved. Although nanoparticle-filled polymers will continue to evolve toward improved properties for materials applications, it is exciting to consider the extent to which synthetic nanoparticle-filled polymer materials can be directed to assemble into hierarchically ordered nanocomposites, as found in nature in the abalone nacre or mother of pearl, a naturally occurring organic/inorganic composite. The mechanical properties displayed by these natural composites are a consequence of assembled substructures that support and reinforce the next, higher level architecture. The construction of such complex structures remains a challenge but will lead to materials with new functionalities. Essential to meeting this challenge is establishing guidelines for process optimization, discovering assembly methods that yield a desired structure, and understanding structure-property relationships to predict the performance of a given architecture. With these in hand, nanoparticle/polymer mixtures hold promise for the fabrication of hierarchically ordered materials that have tailored structures and functionalities that span multiple length scales and dimensions.

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Rapid Temporal Reversal in Predator-Driven Natural Selection

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Because of its potentially epochal scope, evolutionary biology is often caricatured as a strictly descriptive science, but recent years have shown that evolution can be studied on short time scales and that evolutionary biology can be both experimental and predictive (*1*, *2*). Here, we report just such an example by demonstrating the occurrence of a predicted reversal in the direction of natural selection on limb length in *Anolis sagrei*, a common Bahamian lizard often found on the ground in the absence of terrestrial predators.

Previous research showed that, when a larger and entirely terrestrial predatory lizard, *Leiocephalus carinatus*, invades, *A. sagrei* becomes more arboreal and that the extent of this habitat shift broadens through time (*3*). Hence, we predicted that the direction of selection operating on limb length in *A. sagrei* would change through time in the presence of *L. carinatus* (*4*): Initially *A. sagrei* occurs mostly on the ground, so individuals with relatively longer legs, being faster (*5*), would be better able to elude the predators and thereby be favored. As *A. sagrei* becomes more arboreal, however, we predicted that selection would favor the reverse because shorter limbs are better suited for movement on the narrow and irregular surfaces *A. sagrei* would use to avoid the terrestrial predator (*5*).

To test this hypothesis, we introduced *L. carinatus* to six small Bahamian islands that naturally contained *A. sagrei*, randomly choosing six others to serve as controls (*L. carinatus* occurs on nearby larger islands and is known to colonize smaller islands); the number of *L. carinatus* introduced (all adults) was proportional to the number of *A. sagrei* resident on the island. Before introduction of *L. carinatus*, *A. sagrei* individuals on each island were measured and individually marked. Islands were exhaustively censused after 6 and 12 months to determine survival (*6*).

All predictions were confirmed. *A. sagrei* on introduction islands became increasingly arboreal, whereas use of the ground remained unchanged in controls (Fig. 1A); differences in perch diameter between lizards on experimental and control islands also increased through time (Fig. 1B). These trends are the same if comparisons are restricted to only those individuals surviving to the end of the experiment, indicating that differences between treatments resulted from individual shifts in behavior, rather than selection among individuals differing in habitat use [see also (*4*)]. In the first 6 months, longer legs were more strongly favored on introduction islands than on controls, but in the second 6 months, selection was reversed and more strongly favored shorter legs on introduction islands [repeated-measures analysis of covariance (ANCOVA), time*treatment interaction, $F_{1,2} = 59.04$, $P = 0.017$ (Fig. 1C)]. This reversal was accompanied by an especially great divergence in perch diameter during the second six months (repeated-measures analysis of variance, time*treatment interaction, $F_{1,10} = 11.38$, $P = 0.007$).

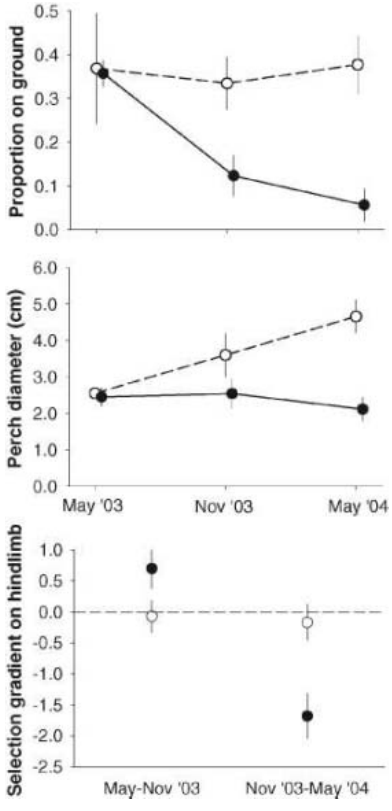

Fig. 1. Changes in habitat use and pattern of natural selection. For use of the ground (**top**) and perch diameter (**middle**), data from May 2003 represents habitat use before the initiation of the experiment. All data are for individuals initially measured and marked in May 2003. Lizards grew throughout the experiment, probably explaining the increase in perch diameter on control islands (an intraspecific relationship between body size and perch diameter is well established in *Anolis* lizards). (**Bottom**) Selection gradients were calculated for two time periods, May 2003 to November 2003 and November 2003 to May 2004. Selection gradients in the figure were adjusted for log-transformed island area (included in the repeated-measures analysis as a covariate) by using least squares means from the ANCOVA. Open symbols indicate control islands; filled symbols, introduction islands. Error bars are ± 1 standard error.

Thus, we showed that selection dramatically changed direction over a very short time, within a single generation, favoring first longer and then shorter hindlimbs. The behavioral shift from the ground to higher perches of smaller diameter apparently caused this remarkable reversal; behavioral flexibility, indeed, may often be the key in driving extremely rapid reversals in evolution (*4*). Our experiment also illustrates the complexity of measuring natural selection in the field (*7*). For example, had we waited 12 months to first measure selection, we could have concluded that the predator's presence had little if any effect on selection, and we certainly would have failed to detect the transitory selection for longer hind limbs over the first 6-month period (table S1).

Nonetheless, over the much longer term, we expect that in the continued presence of *L. carinatus* the initially long-legged *A. sagrei* would remain on high, thin perches, eventually evolving substantially shorter legs. We base this expectation both on functional studies of locomotion (*5*) and on the observation that twig specialists have arisen four times on Caribbean islands, in each case evolving very short hind legs (*8*). Evolutionary biology is by its nature an historical science, but the combination of microevolutionary experimentation and macroevolutionary historical analysis can provide a rich understanding about the genesis of biological diversity.

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Supporting Online Material

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Materials and Methods
Fig. S1
Table S1

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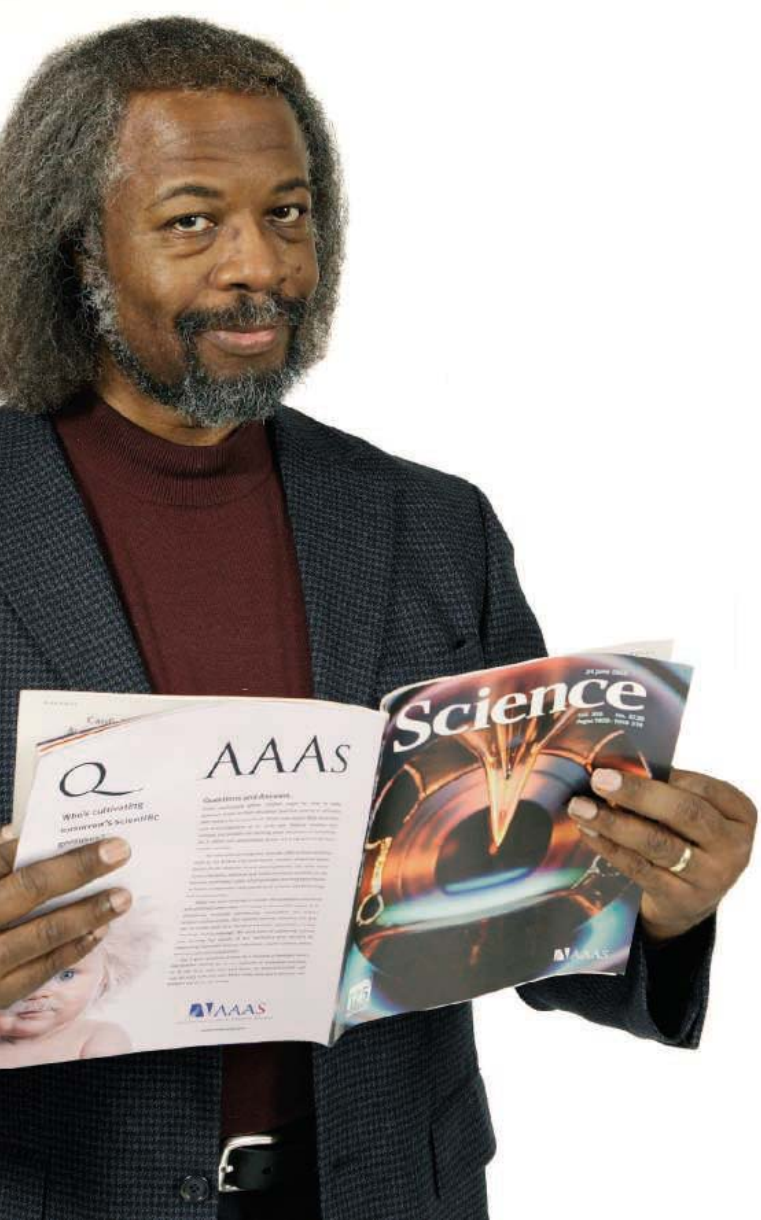

"As a child I got very interested in space travel. When I was six my father gave me some books on rockets and stars. And my universe suddenly exploded in size because I realized those lights in the sky I was looking at were actually places.

I wanted to go there. And I discovered that science and technology was a gift that made this possible. The thrill of most Christmas presents can quickly wear off. But I've found that physics is a gift that is ALWAYS exciting.

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Sequencing and Analysis of Neanderthal Genomic DNA

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Our knowledge of Neanderthals is based on a limited number of remains and artifacts from which we must make inferences about their biology, behavior, and relationship to ourselves. Here, we describe the characterization of these extinct hominids from a new perspective, based on the development of a Neanderthal metagenomic library and its high-throughput sequencing and analysis. Several lines of evidence indicate that the 65,250 base pairs of hominid sequence so far identified in the library are of Neanderthal origin, the strongest being the ascertainment of sequence identities between Neanderthal and chimpanzee at sites where the human genomic sequence is different. These results enabled us to calculate the human-Neanderthal divergence time based on multiple randomly distributed autosomal loci. Our analyses suggest that on average the Neanderthal genomic sequence we obtained and the reference human genome sequence share a most recent common ancestor ~706,000 years ago, and that the human and Neanderthal ancestral populations split ~370,000 years ago, before the emergence of anatomically modern humans. Our finding that the Neanderthal and human genomes are at least 99.5% identical led us to develop and successfully implement a targeted method for recovering specific ancient DNA sequences from metagenomic libraries. This initial analysis of the Neanderthal genome advances our understanding of the evolutionary relationship of *Homo sapiens* and *Homo neanderthalensis* and signifies the dawn of Neanderthal genomics.

Neanderthals are the closest hominid relatives of modern humans (*1*). As late as 30,000 years ago, humans and Neanderthals coexisted in Europe and western Asia (*2*). Since that time, our species has spread across Earth, far surpassing any previous hominid or primate species in numbers, technological development, and environmental impact, while Neanderthals have vanished. Molecular studies of Neanderthals have been exclusively constrained to the comparison of human and polymerase chain reaction (PCR)–amplified Neanderthal mitochondrial sequences, which suggest that the most recent common ancestor of humans and Neanderthals existed ~500,000 years ago, well before the emergence of modern humans (*3*–*5*). Further analyses of mitochondrial data, including the comparison of mitochondrial sequences obtained from several Neanderthals and early modern humans, suggest little or no admixture between Neanderthal and modern human populations in Europe (*3*, *4*, *6*, *7*). However, a major limitation of all prior molecular studies of Neanderthals is that mitochondrial sequences reflect only maternal inheritance of a single locus. Accordingly, in the absence of Neanderthal autosomal and Y-chromosome sequences, the assessment of human-Neanderthal admixture remains incomplete. Mitochondrial data also provide no access to the gene and gene regulatory sequence differences between humans and Neanderthals that would help to reveal biological features unique to each. These insights await the recovery of Neanderthal genomic sequences.

The introduction of high-throughput sequencing technologies and recent advances in metagenomic analysis of complex DNA mixtures now provide a strategy to recover genomic sequences from ancient remains (*8*–*11*). In contrast to previous efforts to obtain ancient sequences by direct analysis of extracts (*3*–*6*, *12*), metagenomic libraries allow the immortalization of DNA isolated from precious ancient samples, obviating the need for repeated destructive extractions (*10*). In addition, once an ancient DNA fragment is cloned into a metagenomic library, it can be distinguished from contamination that might be introduced during subsequent PCR amplification or sequencing by the vector sequences linked to each library-derived insert (Fig. 1).

Recovery of Neanderthal nuclear DNA sequences using a metagenomic approach. In this study, we applied an amplification-independent direct cloning method to construct a Neanderthal metagenomic library, designated NE1, using DNA extracted from a 38,000-year-old specimen from Vindija, Croatia (*6*, *13*). We have recovered 65,250 base pairs (bp) of Neanderthal genome sequence from this library through a combination of Sanger sequencing and massively parallel pyrosequencing. We have also used the metagenomic library as a substrate to isolate specific Neanderthal sequences by direct genomic selection. Several lines of evidence indicated that the hominid sequences in this library were largely Neanderthal, rather than modern human contamination. Mitochondrial PCR analysis of the extract used to build the library, using an amplicon of similar size as the average hominid sequence identified in the library, revealed that only ~2% of the products were from contaminating modern human DNA, whereas the remaining 98% were Neanderthal. Signatures of damage in the hominid sequences that are characteristic of ancient DNA also suggested that they were ancient. Finally and most importantly, comparison of hominid sequences from the library to orthologous human and chimpanzee genomic sequences identified human-specific substitutions at sites where the hominid sequence was identical to that of the chimpanzee, enabling us to make estimates of the human-Neanderthal divergence time (*3*, *4*, *6*).

We initially assessed the Neanderthal genomic sequence content of library NE1 by Sanger sequencing of individual clones, which allowed individual library inserts to be completely sequenced and thus provided a direct measure of hominid insert size that could not be obtained from the ~100-bp pyrosequencing reads described below (Table 1). We identified hominid sequences in the library by

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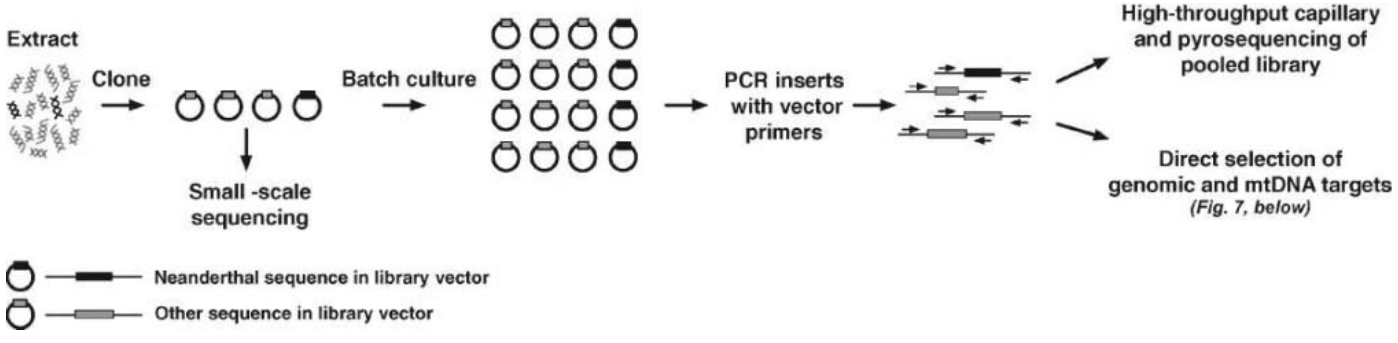

Fig. 1. Generation of ancient metagenomic library DNAs for direct selection and pyrosequencing.

BLAST comparison to the reference human genome sequence (*13*, *14*). In many cases, the human BLAST hit covered only part of the insert, because the direct cloning method we employed produces chimeric inserts consisting of smaller fragments ligated into larger concatemers. The small average size of these putatively ancient Neanderthal fragments (52 bp) is similar to results we previously obtained from two Pleistocene cave bear libraries, in which the average library insert size was between 100 and 200 bp, whereas BLAST hits to reference carnivore genome sequences were on average 69 bp (Fig. 2) (*10*). The small BLAST hit sizes and insert sizes in both cave bear and Neanderthal metagenomic libraries are consistent with the degradation of ancient genomic DNA into small fragments over tens of thousands of years, illustrating the general condition of nuclear DNA in ancient remains.

Sanger sequencing of individual clones from library NE1 suggested that it contained sufficient amounts of Neanderthal sequence to conduct a random sequence survey of the Neanderthal genome. However, the small percentage of clones we identified as containing hominid sequences indicated that we would have to sequence a very large number of clones to obtain enough Neanderthal genome sequence for this analysis. We therefore carried out deep sequencing of pooled inserts from library NE1 using massively parallel pyrosequencing. To obtain pooled inserts, we amplified transformed NE1 library DNA in liquid batch culture and recovered library inserts from purified plasmid DNA by PCR (Fig. 1). We generated 1.47 million pyrosequencing reads, compared each to the human genome sequence with MEGABLAST, and obtained 7880 hits. Assembly of these reads and reanalysis of the resulting scaffolds by BLASTN produced 1126 unique Neanderthal loci, yielding 54,302 bp of Neanderthal genomic sequence (*13*).

Assessment of pyrosequencing data quality by comparison to Sanger sequence data. The pyrosequencing approach generates significant amounts of sequence but does so with a higher error rate than Sanger sequencing (*11*). To assess the quality of Neanderthal pyrosequencing data, we generated consensus sequences from pyrosequencing reads overlapping the same Neanderthal genomic locus and filtered out low-quality positions in the resulting contigs (quality score < 15). To determine whether these contigs contained additional errors not detectable by quality-score filtering, we also used Sanger sequencing to analyze 19,200 clones from the same batch culture used to generate the pyrosequencing data. This sequencing yielded 130 loci (6.2 kb) that were also represented in the pyrosequencing data. Sanger sequencing and pyrosequencing results for these 130 Neanderthal loci agreed at 99.89% of ungapped positions. In addition, Sanger sequencing and pyrosequencing yielded Neanderthal sequences that were nearly equally divergent from the human reference sequence (pyrosequencing = 0.47% divergence, Sanger sequencing = 0.49%). These results indicate that the frequency of single-base errors is probably no greater in Neanderthal genomic sequence obtained from assembled quality-filtered pyrosequencing data than in sequence obtained from Sanger sequencing.

The low complexity of library NE1 made these analyses possible, because it resulted in a limited number of clones in the library that were amplified by batch culture and PCR and then sequenced in depth (fig. S1). We estimated that the coverage obtained in library NE1 (~0.002%) is significantly lower than that previously obtained in cave bear metagenomic libraries prepared from samples of similar age as the Neanderthal sample used here (*10*). The low coverage in library NE1 is more likely due to the quality of this particular library rather than being a general feature of ancient DNA. Nevertheless, we were able to obtain substantial amounts of authentic Neanderthal genomic sequence from the library by deep sequencing.

Comparison of orthologous Neanderthal, human, and chimpanzee genomic sequences. To ascertain whether the library NE1 hominid sequence we obtained was a representative sampling of the Neanderthal genome, we identified each NE1 library sequence for which the bit score of the best BLASTN hit in the human genome was higher than the bit scores of all other hits for that sequence. We then determined the distribution of all such best BLASTN hits across human chromosomes [43,946 bp in 1,039 loci (table S1 and Fig. 3A)]. The amount of Neanderthal sequence aligned to each human chromosome was highly correlated with sequenced chromosome length, indicating that the Neanderthal sequences we obtained were randomly drawn from all chromosomes (Pearson correlation coefficient = 0.904, Fig. 3A). The hominid hits included Y-chromosome sequences, demonstrating that our sample was derived from a Neanderthal male. We annotated each Neanderthal locus according to the annotations (known genes, conserved noncoding sequences, and repeats) associated with the aligned human sequence (table S2). Neanderthal sequences obtained by both Sanger sequencing and pyrosequencing showed a distribution of sequence features consistent with the known distribution of these features in the human genome (Fig. 3B). These sequences are therefore likely to represent a random sampling of the Neanderthal genome.

Comparison of authentic Neanderthal sequence with orthologous human and chimpanzee genomic sequences will reveal sites at which Neanderthal is identical to chimpanzee but at which the human sequence has undergone a mutation since the human-Neanderthal divergence. Determining the number of human-specific mutations is critical to dating the human-Neanderthal split. To identify these events, we constructed alignments of orthologous human, Neanderthal, and chimpanzee sequences and identified mutations specific to each lineage by parsimony (*15*). We identified 34 human-specific substitutions in 37,636 human, Neanderthal, and chimpanzee aligned positions, including substitutions on chromosomes X and Y that were not considered in subsequent analyses.

We also identified 171 sites with Neanderthal-specific substitutions relative to human and chimpanzee. It has been shown that nucleotides in genuine ancient DNA are occasionally chemically damaged, most frequently because of the deamination of cytosine to uracil, resulting in the incorporation of incorrect bases during PCR and sequencing (*16*). This results in an apparent excess of C-to-T and G-to-A mismatches (which are equivalent events) between the ancient sequence and the modern genomic reference sequence. We observe a significant excess of C-to-T and G-to-A mismatches (relative to T-to-C and A-to-G mismatches) between human and NE1 hominid sequences obtained by both Sanger sequencing and pyrosequencing [$P \ll 0.0005$, Fisher's exact test (Fig. 4 and table S3)]. This accounts for the large number of Neanderthal-specific substitutions we observe and further supports the supposition that the hominid sequences are Neanderthal in origin. Despite the bias toward C-to-T and G-to-A events in Neanderthal genomic sequence, the overall frequency of these events is

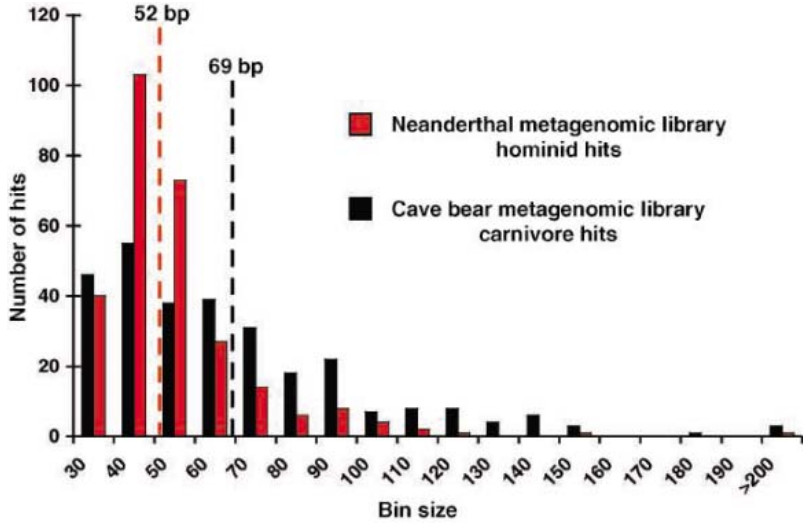

Fig. 2. Size distribution, plotted in 10-bp bins, of Neanderthal and cave bear sequences obtained from metagenomic libraries by Sanger sequencing of individual clones. The average hit size in each case is indicated by a dotted line.

low (~0.37% of all sites), indicating that the vast majority of human-Neanderthal-chimpanzee aligned positions are not likely to be significantly affected by misincorporation errors (*13*).

The length distribution of ancient DNA fragments shown in Fig. 2, when combined with the sequence signatures of ancient DNA described above, offers another metric for assessing the degree of modern human contamination in our library. Based on the assumption that modern contaminating DNA fragments would be longer than authentic ancient DNAs, which is supported by the observation that contaminating modern human DNA fragments in the cave bear libraries were on average much longer than the cave bear sequences (116 versus 69 bp) (*10*), we examined the distribution of human-Neanderthal mismatches in our data set as a function of alignment length. If a substantial fraction of the hominid sequence recovered from the Neanderthal sample were actually modern human DNA, we would expect to see a lower human-Neanderthal sequence divergence in the longer BLASTN alignments than we observe in the entire data set, because the longer hominid sequences would be enriched in modern human contaminants. The excess of damage-induced Neanderthal-specific mismatches described above would also be expected to decrease as alignment length increases, because individual bases in the longer modern human fragments would show relatively few chemical modifications. However, we did not observe these trends in our Neanderthal sequence. The human-Neanderthal sequence divergence in all autosomal alignments greater than 52 bp (the approximate midpoint of the distribution shown in Fig. 2) was similar to the divergence obtained from the whole data set (0.59% versus 0.52%). The excess of C-to-T and G-to-A mismatches was also maintained in the longer alignments. These results further support the supposition that the hominid sequence we obtained is predominantly Neanderthal in origin.

Coalescence time of human and Neanderthal genomic sequences. These data allowed us to examine for the first time the genetic relationship between humans and Neanderthals using nuclear genomic sequences (*13*). We first considered the average coalescence time for the autosomes between the Neanderthal genomic sequence that we obtained and the reference human genome sequence. We observed 502 human-chimpanzee autosomal differences in the human-Neanderthal-chimpanzee sequence alignments we constructed. Based on comparison to the Neanderthal sequence, 27 of these differences were human-specific and therefore postdate the most recent common ancestor (MRCA) of the human and Neanderthal sequences. Using this information, our maximum likelihood estimate of the average time to the MRCA of these sequences is 706,000 years, with a 95% confidence interval (CI) of 468,000 to 1,015,000 years (Figs. 5A and 6) (*13*). This calculation does not make use of Neanderthal-specific changes, because many of those events are due to DNA damage as described above. In addition, we restricted our analysis to autosomal data, because these represent 97% of our total data set and population genetic parameters are likely to differ between the autosomes and sex chromosomes. Our estimate uses a mutation rate obtained by setting the average coalescence time for human and chimpanzee autosomes to 6.5 million years ago, a value that falls within the range suggested by recent studies (*17*, *18*). Inaccuracies in the human-chimpanzee divergence time would shift all the time estimates and CIs presented here in proportion to the error.

Split time of ancestral human and Neanderthal populations. Our estimate of the average common ancestor time reflects the average time at which the Neanderthal and human reference sequences began to diverge in the common ancestral population, not the actual split time of the ancestral populations that gave rise to Neanderthals and modern humans. To estimate the actual split time of the ancestral human and Neanderthal populations, we developed a method that incorporated data from the human and Neanderthal reference sequences, as well as genotypes from 210 individuals with genome-wide single-nucleotide polymorphism (SNP) data collected by the International HapMap Consortium (Table 2) (*19*). We included the HapMap data because they indicate what proportion of sites in the Neanderthal sequence fall within the spectrum of modern human variation. For example, if the ancestral human and Neanderthal populations split long ago, before the rise of most modern human genetic diversity captured by the HapMap data, then Neanderthal sequence would almost never carry the derived allele, relative to the orthologous chimpanzee sequence, for a human SNP (Table 2). Conversely, a more recent population split would result in Neanderthal sequence frequently carrying the derived allele for human SNPs.

To formalize this idea, we considered an explicit population model for the relationship between Neanderthals and each HapMap population (East Asians,

Table 1. Amount of unique Neanderthal sequence obtained from library NE1 by Sanger sequencing of individual clones, as well as Sanger sequencing and pyrosequencing of clones in batch culture. n.a., not applicable.

	Individual clones	Batch culture	
Sequencing chemistry	Sanger	Sanger	Pyrosequencing
Reads	9984	19,200	1,474,910
Average insert	134 bp	196 bp	n.a.
Average BLAST hit	52 bp	52 bp	48 bp
Unique loci	131	69	1126
Total unique hominid sequence	6845 bp	4103 bp	54,302 bp

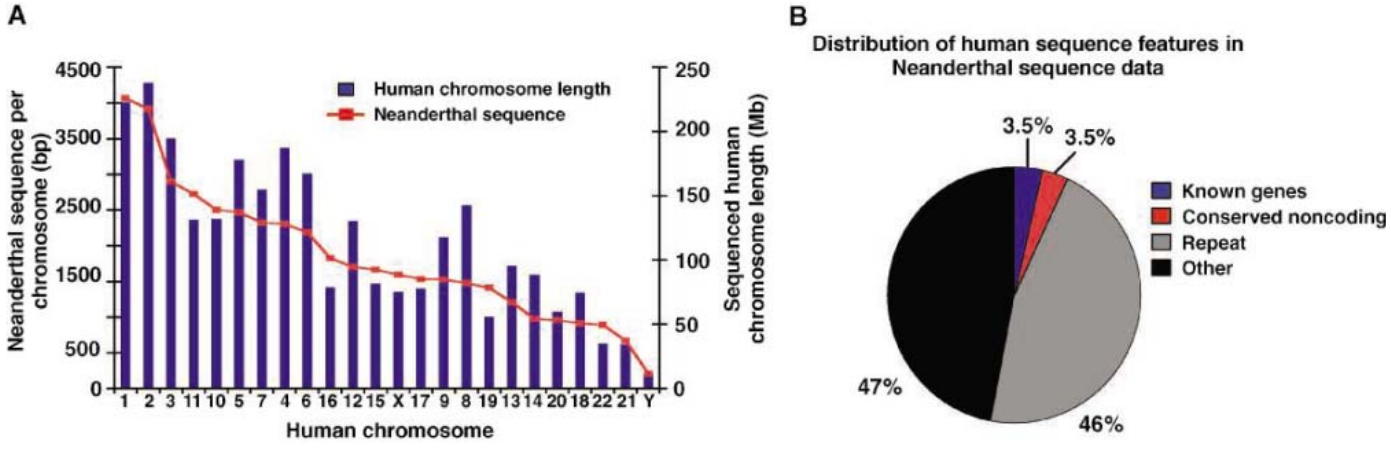

Fig. 3. (**A**) Representation of each Neanderthal chromosome in 43.9 kb of NE1 hominid sequences displaying a statistically unambiguous best BLAST hit to the human genome, relative to the total sequenced length of each human chromosome minus gaps. Chromosomes are ranked by the amount of Neanderthal sequence aligned to each. Chromosomes X and Y are shown at half their total length to correct for their haploid state in males relative to the autosomes. (**B**) Representation of sequence features in the NE1 hominid sequence shown in (A).

Europeans, and Yoruba) separately (fig. S3) (*13*). We assumed that Neanderthals and modern humans evolved from a single ancestral population of 10,000 individuals and that the Neanderthal population split away from the human ancestral population instantaneously at a time T in the past, with no subsequent gene flow. In order to model the demographic histories of the HapMap populations, we made use of models and parameters estimated by Voight *et al.* (*20*) based on resequencing data from 50 unlinked, noncoding regions. Those demographic models include bottlenecks for East Asians and Europeans and modest exponential growth for Yoruba (*13*).

We then constructed a simulation-based composite likelihood framework to estimate the time of the human-Neanderthal population split (*13*, *21*). At each site in the human-Neanderthal-chimpanzee alignments we constructed, we recorded the Neanderthal and human reference alleles relative to chimpanzee. We also determined, separately for each population, whether each site was a HapMap SNP in that population and if so, the allele frequency (Table 2).

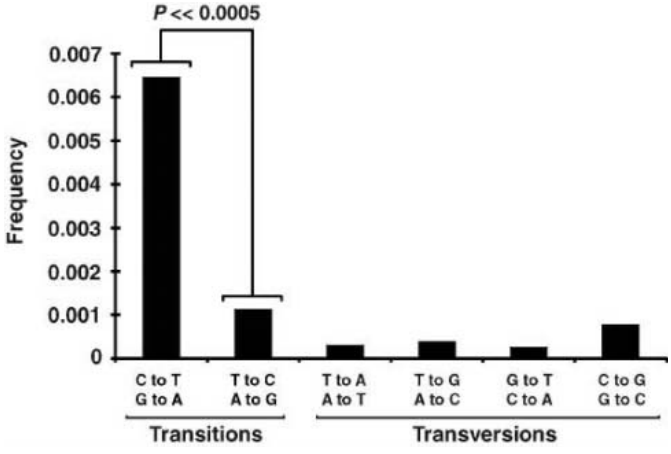

Fig. 4. Frequency distribution of 171 Neanderthal-specific substitutions observed in 37,636 bp of aligned human, Neanderthal, and chimpanzee genomic sequence. Complementary substitutions (such as C to T and G to A) are considered equivalent events.

We used simulations to estimate the probability of each possible data configuration at a single site as a function of the human-Neanderthal split time. The simulations used the estimated population demography for each HapMap population and a probabilistic model of SNP ascertainment to match the overall density and frequency spectrum of HapMap Phase II SNPs. Likelihood curves for the split time were computed by multiplying likelihoods across sites as though they were independent. In practice, this is an excellent approximation for our data because the Neanderthal sequence reads are very short and just 1 out of 905 aligned fragments contains more than one human-specific allele or SNP. Bootstrap simulations confirmed that our composite likelihood method yields appropriate CIs for the split time (*13*).

Using this approach, the maximum likelihood estimates for the split time of the ancestral human and Neanderthal populations are 440,000 years (95% CI of 170,000 to 620,000 years) based on the European data, 390,000 years (170,000 to 670,000 years) for East Asians, and 290,000 years (120,000 to 570,000 years) for Yoruba (Figs. 5B and 6). These values predate the earliest known appearance of anatomically modern humans in Africa ~195,000 years ago (*22*). Because these split times are before the migration of modern humans out of Africa, the three population-specific estimates should

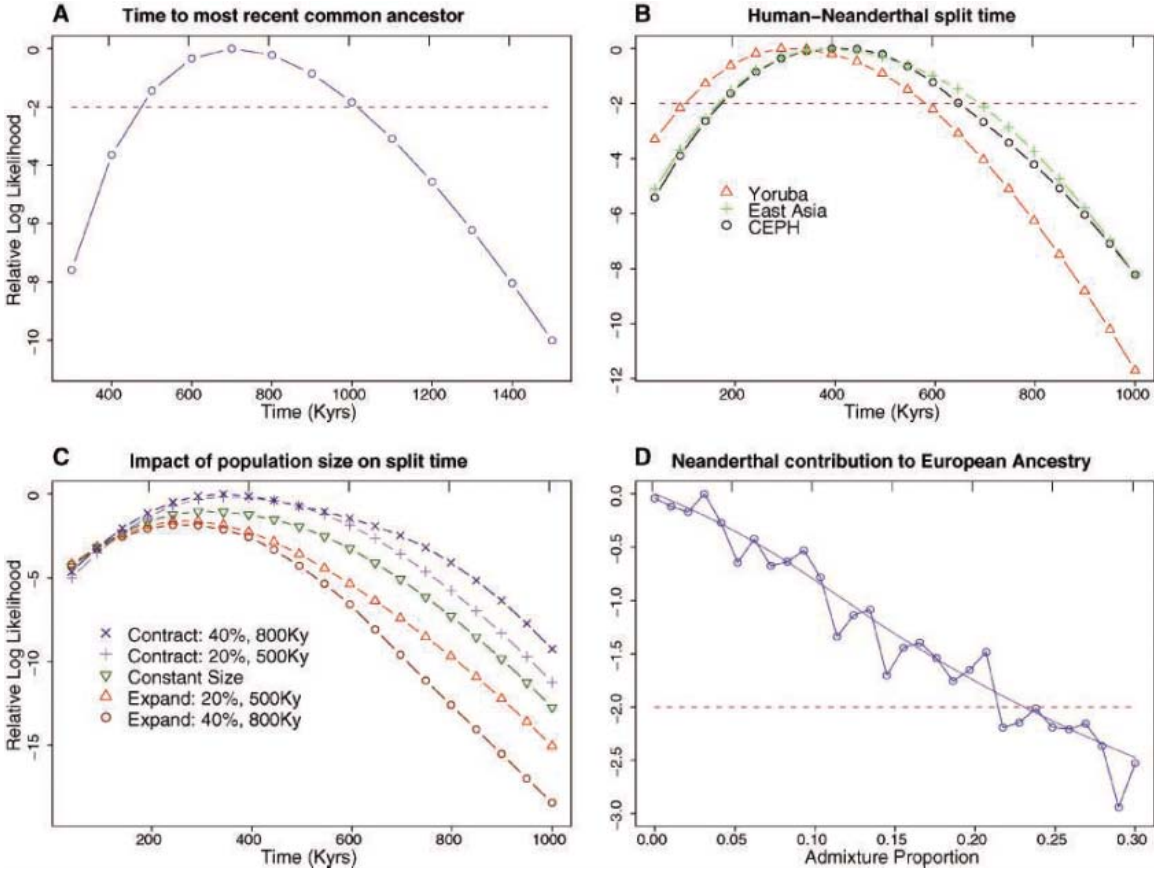

Fig. 5. (**A**) Log-likelihood curve of the time to the MRCA of the Neanderthal and human reference sequences. (**B**) Smoothed relative log-likelihood estimates of the split times between different human populations and the Neanderthal population. (**C**) Impact of changes in the ancient population size on split time estimates for five models that are consistent with modern polymorphism data. Ky, thousand years. Each curve is the smoothed log likelihood relative to the maximum over all five models. For each model, the text on the plot indicates the degree of expansion or contraction and the time before the present at which the size change occurred. The expansion models are less likely as compared to either constant population size or the contraction models. (**D**) The log-likelihood estimates of the contribution of the Neanderthal population to the ancestry of Europeans. The light blue line is a smoothed version of the estimates. The horizontal dashed maroon line in (A), (B), and (D) represents a 2 log-likelihood drop, and the region bounded by this line represents the 95% CI around the maximum likelihood estimates.

all be estimates of the same actual split time. The average of these estimates, ~370,000 years, is thus a sensible point estimate for the split time. Substantial contamination with modern human DNA would cause these estimates to be artificially low, but 2% contamination, the rate suggested by mitochondrial PCR analysis of the primary extract used to construct the library, would have essentially no impact (*13*).

Our estimates of the human-Neanderthal split time might depend heavily on the assumption that the ancestral effective population size of humans was 10,000 individuals. To address this, we explored a set of models in which the ancestral human population expanded or contracted at least 200,000 years ago (*13*). We found that much of the parameter space—though not the original model—could be excluded on the basis of modern human polymorphism data from Voight *et al*. (*20*). We repeated our likelihood analysis of the Neanderthal data using models incorporating ancient expansion or contraction that are consistent with modern data and found that these did not substantially change our population split time estimates (Fig. 5C).

Our data include three sites at which Neanderthal carries the derived allele for a polymorphic HapMap SNP. These sites are unlikely to represent modern contamination because for two of the SNPs, the derived allele is found only in Yoruba; also, one of the SNPs lies on a fragment that contains a C-to-T transition in Neanderthals that is characteristic of chemical damage to DNA. These observations indicate that the Neanderthal sequence may often coalesce within the human ancestral tree. Based on simulations of our best-fit model for Yoruba, we estimate that Neanderthal is a true outgroup for approximately 14% (assuming a split time of 290,000 years, the Yoruba estimate) to 26% (assuming a split time of 440,000 years, the European estimate) of the autosomal genome of modern humans, although more data will be required to achieve a precise estimate.

Lack of evidence for admixture between humans and Neanderthals. Because Neanderthals coexisted with modern humans in Europe, there has long been interest in whether Neanderthals might have contributed to the European gene pool. Previous studies comparing human and Neanderthal mitochondrial sequences did not find evidence of a Neanderthal genetic contribution to modern humans. However, the utility of mitochondrial data in addressing this question is limited in that it is restricted to a single locus and, due to the maternal inheritance of mitochondrial DNA, is informative only about admixture between Neanderthal females and modern human males (*3*–*6*). Moreover, it has been argued that some aspects of modern human autosomal data may be the result of modest levels of Neanderthal admixture (*23*).

If Neanderthal admixture did indeed occur, then this could manifest in our data as an abundance of low-frequency derived alleles in Europeans where the derived allele matches Neanderthal. No site in the data set appears to be of this type. In order to formally evaluate this hypothesis, we extended our composite likelihood simulations to include a single admixture event 40,000 years ago in which a fraction p of the European gene pool was derived from Neanderthals. We fixed the human-Neanderthal split at 440,000 years ago (the split time estimate for Europeans). With these assumptions, the maximum likelihood estimate for the Neanderthal contribution to modern genetic diversity is zero. However, the 95% CI for this estimate ranges from 0 to 20%, so a definitive answer to the admixture question will require additional Neanderthal sequence data (Fig. 5D).

Targeted recovery of specific Neanderthal sequences by direct genomic selection. Although we have recovered significant amounts of Neanderthal genome sequence using a metagenomic approach, hundreds of gigabases of sequence would be required to achieve reasonable coverage of a single Neanderthal genome by this method. Moreover, our results indicate that at least 99.5% of the Neanderthal sequence that would be obtained would be identical to the modern human sequence. The human-Neanderthal sequence differences that would yield great insight into human biology and evolution are thus rare events in an overwhelming background of uninformative sequence. We therefore explored the potential of metagenomic libraries to serve as substrates to recover specific Neanderthal sequences of interest by targeted methods. To this end, we developed a direct genomic selection approach to recover known and unknown sequences from metagenomic ancient DNA libraries (Fig. 7) (*24*). We first attempted to recover specific sequences from a Pleistocene cave bear metagenomic library we previously constructed. We designed PCR probes corresponding to 96 sequences highly conserved among mammals but not previously shown to be present in the cave bear library. We amplified these sequences from the human genome and hybridized the resulting probes to PCR-amplified cave bear library inserts produced as described above (Fig. 1). Recovered library DNAs were amplified by PCR and sequenced. We successfully recovered five targets consisting of a known enhancer of *Sox9* and conserved sequences near *Tbx3*, *Shh*, *Msx2*, and *Gdf6* (table S4). In principle, these sequences could be derived from contaminating DNA rather than the cave bear library. Critically, the captured cave bear sequences were flanked by library vector sequence, directly demonstrating that these sequences were derived from a cloned library insert and not from contaminating DNA introduced during direct selection (Fig. 7 and fig. S2).

Based on these results, we attempted to recover specific Neanderthal sequences from library NE1. We focused on recovering sequences that we had previously identified by shotgun sequencing because of the low complexity of library NE1, and were able to recover 29 of 35 sequences we targeted (table S4). The authenticity of these sequences was confirmed by the presence of library vector sequences in the reads. Our

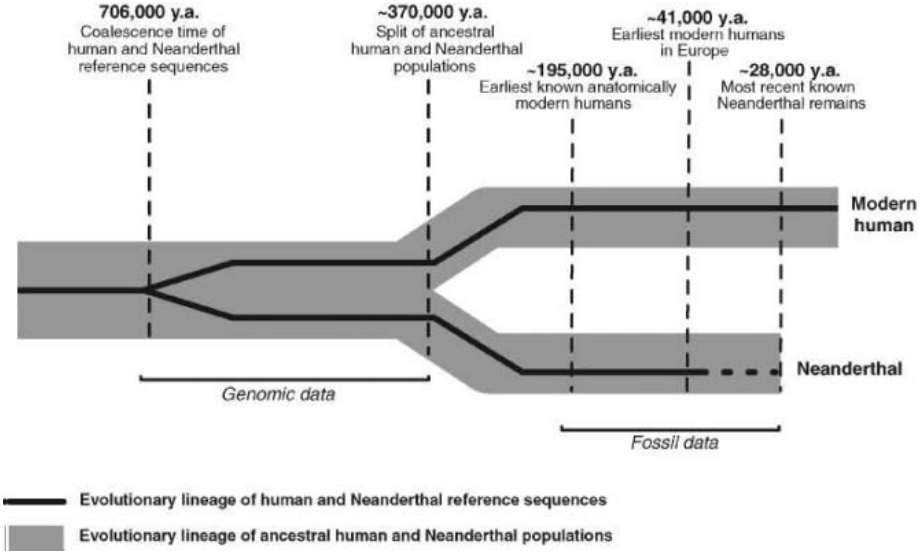

Fig. 6. Divergence estimates for human and Neanderthal genomic sequences and ancestral human and Neanderthal populations, shown relative to dates of critical events in modern human and Neanderthal evolution (*2*, *22*, *25*). The branch lengths are schematic and not to scale. y.a., years ago.

Table 2. Summary of all autosomal sites sequenced in Neanderthal and uniquely aligned to the human and chimpanzee reference sequences. The designations "ancestral" and "derived" indicate whether each site is, respectively, a match or mismatch with chimpanzee. Sites are partitioned into those that overlap a Phase II HapMap SNP (with SNPs) and those that do not (without SNPs).

		Sequence state in human reference	
	With SNPs	Ancestral	Derived
Sequence state in Neanderthal	Ancestral	24	8
	Derived	3	0
		Sequence state in human reference	
	Without SNPs	Ancestral	Derived
Sequence state in Neanderthal	Ancestral	35,801	19
	Derived	161	475

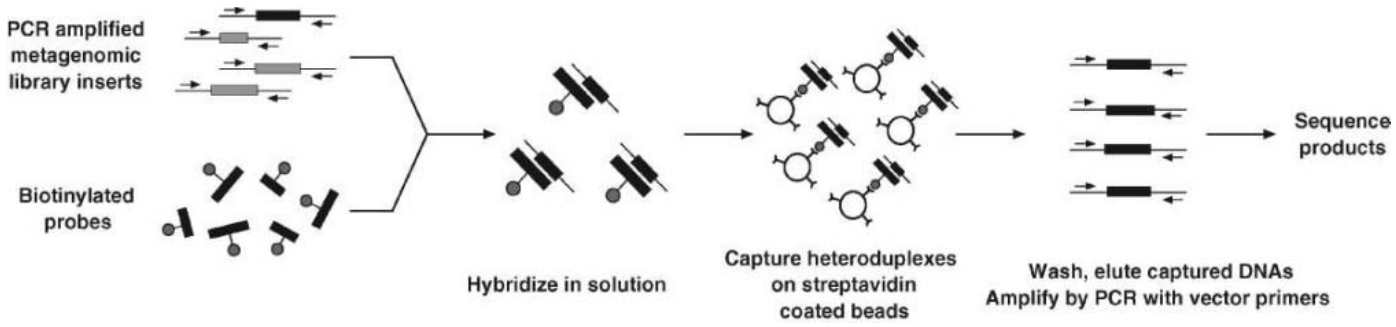

Fig. 7. Recovery of Neanderthal genomic sequences from library NE1 by direct genomic selection.

success in recovering both previously unknown cave bear and known Neanderthal genomic sequences using direct genomic selection indicates that this is a feasible strategy for purifying specific cloned Neanderthal sequences out of a high background of Neanderthal and contaminating microbial DNA. This raises the possibility that, should multiple Neanderthal metagenomic libraries be constructed from independent samples, direct selection could be used to recover Neanderthal sequences from several individuals to obtain and confirm important human-specific and Neanderthal-specific substitutions.

Conclusions. The current state of our knowledge concerning Neanderthals and their relationship to modern humans is largely inference and speculation based on archaeological data and a limited number of hominid remains. In this study, we have demonstrated that Neanderthal genomic sequences can be recovered using a metagenomic library-based approach and that specific Neanderthal sequences can be obtained from such libraries by direct selection. Our study thus provides a framework for the rapid recovery of Neanderthal sequences of interest from multiple independent specimens, without the need for whole-genome resequencing. Such a collection of targeted Neanderthal sequences would be of immense value for understanding human and Neanderthal biology and evolution. Future Neanderthal genomic studies, including targeted and whole-genome shotgun sequencing, will provide insight into the profound phenotypic divergence of humans both from the great apes and from our extinct hominid relatives, and will allow us to explore aspects of Neanderthal biology not evident from artifacts and fossils.

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26. Neanderthal sequences reported in this study have been deposited in GenBank under accession numbers DX935178 to DX936503. We thank E. Green, M. Lovett, and members of the Rubin, Pääbo, and Pritchard laboratories for insightful discussions and support. J.P.N. was supported by NIH National Research Service Award fellowship 1-F32-GM074367. G.C. and S.K. were supported by grant R01 HG002772-1 (NIH) to J.K.P. This work was supported by grant HL066681, NIH Programs for Genomic Applications, funded by the National Heart, Lung and Blood Institute; and by the Director, Office of Science, Office of Basic Energy Sciences, of the U.S. Department of Energy under contract number DE-AC02-05CH11231.

Supporting Online Material

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Materials and Methods
Figs. S1 to S6
Tables S1 to S12
References

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REPORTS

Resilient Machines Through Continuous Self-Modeling

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Animals sustain the ability to operate after injury by creating qualitatively different compensatory behaviors. Although such robustness would be desirable in engineered systems, most machines fail in the face of unexpected damage. We describe a robot that can recover from such change autonomously, through continuous self-modeling. A four-legged machine uses actuation-sensation relationships to indirectly infer its own structure, and it then uses this self-model to generate forward locomotion. When a leg part is removed, it adapts the self-models, leading to the generation of alternative gaits. This concept may help develop more robust machines and shed light on self-modeling in animals.

Robotic systems are of growing interest because of their many practical applications as well as their ability to help understand human and animal behavior (*1*–*3*), cognition (*4*–*6*), and physical performance (*7*). Although industrial robots have long been used for repetitive tasks in structured environments, one of the long-standing challenges is achieving robust performance under uncertainty (*8*). Most robotic systems use a manually constructed mathematical model that captures the robot's dynamics and is then used to plan actions (*9*). Although some parametric identification methods exist for automatically improving these models (*10*–*12*), making accurate models is difficult for complex machines, especially when trying to account for possible topological changes to the body, such as changes resulting from damage.

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Although much progress has been made in allowing robotic systems to model their environment autonomously (*8*), relatively little is known about how a robot can learn its own morphology, which cannot be inferred by direct observation or retrieved from a database of past experiences (*13*). Without internal models, robotic systems can autonomously synthesize increasingly complex behaviors (*6*, *14*–*16*) or recover from damage (*17*) through physical trial and error, but this requires hundreds or thousands of tests on the physical machine and is generally too slow, energetically costly, or risky.

Here, we describe an active process that allows a machine to sustain performance through an autonomous and continuous process of self-modeling. A robot is able to indirectly infer its own morphology through self-directed exploration and then use the resulting self-models to synthesize new behaviors. If the robot's topology unexpectedly changes, the same process restructures its internal self-models, leading to the generation of qualitatively different, compensatory behavior. In essence, the process enables the robot to continuously diagnose and recover from damage. Unlike other approaches to damage recovery, the concept introduced here does not presuppose built-in redundancy (*18*, *19*), dedicated sensor arrays, or contingency plans designed for anticipated failures (*20*). Instead, our approach is based on the concept of multiple competing internal models and generation of actions to maximize disagreement between predictions of these models.

The process is composed of three algorithmic components that are executed continuously by the physical robot while moving or at rest (Fig. 1): Modeling, testing, and prediction. Initially, the robot performs an arbitrary motor action and records the resulting sensory data (Fig. 1A). The model synthesis component (Fig. 1B) then synthesizes a set of 15 candidate self-models using stochastic optimization to explain the observed sensory-actuation causal relationship. The action synthesis component (Fig. 1C) then uses these models to find a new action most likely to elicit the most information from the robot. This is accomplished by searching for the actuation pattern that, when executed on each of the candidate self-models, causes the most disagreement across the predicted sensor signals (*21*–*24*). This new action is performed by the physical robot (Fig. 1A), and the model synthesis component now reiterates with more available information for assessing model quality. After 16 cycles of this process have terminated, the most accurate model is used by the behavior synthesis component to create a desired behavior (Fig. 1D) that can then be executed by the robot (Fig. 1E). If the robot detects unexpected sensor-motor patterns or an external signal as a result of unanticipated morphological change, the robot reinitiates the alternating cycle of modeling and exploratory actions to produce new models reflecting the change. The new most accurate model is now used to generate a new, compensating behavior to recover functionality. A complete sample experiment is shown in Fig. 2.

We tested the proposed process on a four-legged physical robot that had eight motorized joints, eight joint angle sensors, and two tilt sensors. The space of possible models comprised any planar topological arrangement of eight limbs, including chains and trees (for examples, see Figs. 1 and 2). After damage occurs, the space of topologies is fixed to the previously inferred morphology, but the size of the limbs can be scaled (Fig. 2, N and O). The space of possible actions comprised desired angles that the motors were commanded to reach (*25*). Many other self-model representations could replace the explicit simulations used here, such as artificial neural or Bayesian networks, and other sensory modalities could be exploited, such as pressure and acceleration (here the joint angle sensors were used only to verify achievement of desired angles, and orientation of the main body was used only for self-model synthesis). Nonetheless, the use of implicit representations such as artificial neural networks—although more biologically plausible than explicit simulation—would make the validation of our theory more challenging, because it would be difficult to assess the correctness of the model (which can be done by visual inspection for explicit simulations). More important, without an explicit representation, it is difficult to reward a model for a task such as forward locomotion (which requires predictions about forward displacement) when the model can only predict orientation data.

The proposed process was compared with two baseline algorithms, both of which use random rather than self-model–driven data acquisition. All three algorithm variants used a similar amount of computational effort (~250,000 internal model simulations) and the same number (16) of physical actions (Table 1). In the first baseline algorithm, 16 random actions were executed by the physical robot (Fig. 1A), and the resulting data were supplied to the model synthesis component for batch training (Fig. 1B). In the second baseline algorithm, the action synthesis component output a random action, rather than searching for one that created disagreement among competing candidate self-models. The actions associated with Fig. 1, A to C,

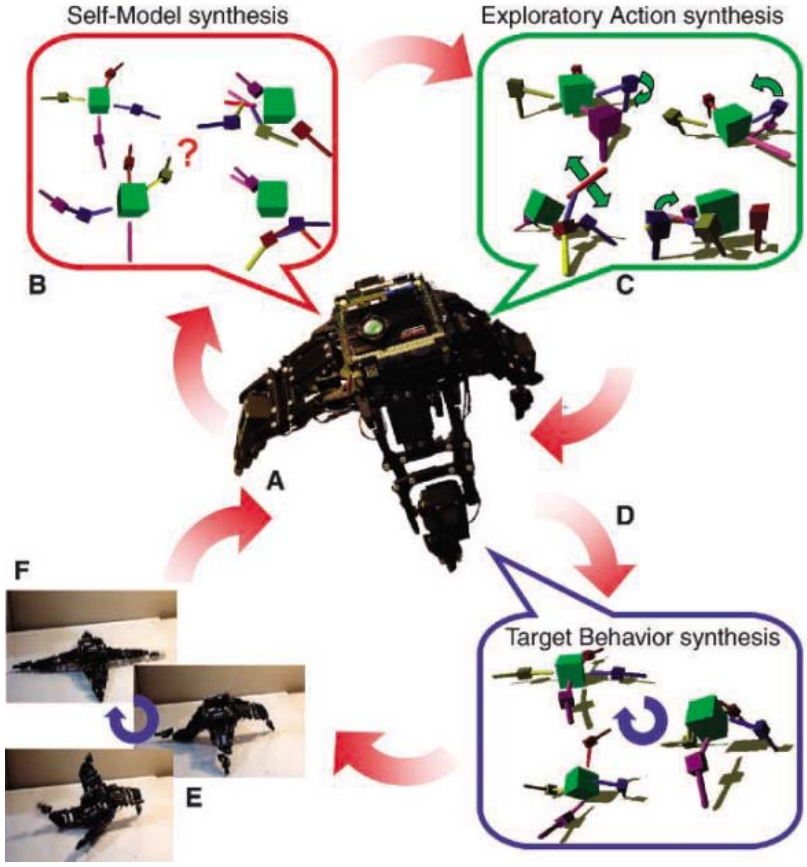

Fig. 1. Outline of the algorithm. The robot continuously cycles through action execution. (**A** and **B**) Self-model synthesis. The robot physically performs an action (A). Initially, this action is random; later, it is the best action found in (C). The robot then generates several self-models to match sensor data collected while performing previous actions (B). It does not know which model is correct. (**C**) Exploratory action synthesis. The robot generates several possible actions that disambiguate competing self-models. (**D**) Target behavior synthesis. After several cycles of (A) to (C), the currently best model is used to generate locomotion sequences through optimization. (**E**) The best locomotion sequence is executed by the physical device. (**F**) The cycle continues at step (B) to further refine models or at step (D) to create new behaviors.

were cycled as in the proposed algorithm, but Fig. 1C output a random action, rather than an optimized one.

Thirty experiments of each of the three algorithm variants were conducted, both before and after the robot suffered damage. Before damage, the robot began each experiment with a set of random models; after damage, the robot began with the best model produced by the model-driven algorithm (Fig. 2F). We found that the probability of inferring a topologically correct model was notably higher for the model-driven algorithm than for either random baseline algorithm (Table 1) and that the final models were more accurate on average in the model-driven algorithm than in either random baseline algorithm (Table 1). Similarly, after damage, the robot was better able to infer that one leg had been reduced in length using the model-driven algorithm than it could using either baseline algorithm (Table 1). This indicates that alternating random actions with modeling, compared with simply performing several actions first and then modeling, does not improve model synthesis (baseline 2 does not outperform baseline 1), but a robot that actively chooses which action to perform next on the basis of its current set of hypothesized self-models has a better chance of successfully inferring its own morphology than a robot that acts randomly (the model-driven algorithm outperforms baseline algorithms 1 and 2).

Because the robot is assumed not to know its own morphology a priori, there is no way for it to determine whether its current models have captured its body structure correctly. We found that disagreement among the current model set (information that is available to the algorithm) is a good indicator of model error (the actual inaccuracy of the model, which is not available to the algorithm), because a positive correlation exists between model disagreement and model error across the $n = 30$ experiments that use the model-driven algorithm (Spearman rank correlation = 0.425, $P < 0.02$). Therefore, the experiment that resulted in the most model agreement (through convergence toward the correct model) was determined to be the most successful from among the 30 experiments performed, and the best model it produced (Fig. 2F) was selected for behavior generation. This was also the starting model that the robot used when it suffered unexpected damage (Table 1).

The behavior synthesis component (Fig. 1D) was executed 30 times with this model, starting each time with a different set of random behaviors. Figure 3 reports the final positions predicted by the model robot using the best behavior produced by each experiment (black dots). Each of those 30 behaviors was then executed on the physical robot, and the resulting actual positions are reported in Fig. 3 (blue dots). As a control, 30 random behaviors were also executed on the physical robot (red dots). Although there is some discrepancy between the predicted distance and actual distance, there is a clear forward motion trend that is absent from the random behaviors. This indicates that this automatically generated self-model was sufficiently predictive to allow the robot to consistently develop forward motion patterns without further physical trials. One of the better locomotion patterns is shown in fig. S1A. The transferal from the self-model to reality was not perfect, although the gaits were qualitatively similar; differences between the simulated and physical gait (seen at 2.6 and 5.2 s) were most likely due to friction and kinematic bifurcations at symmetrical postures, both difficult to predict. Similarly, after damage, the robot was able to synthesize sufficiently accurate models (an example is given in Fig. 2O) for generating new, compensating behaviors that enabled it to continue moving forward. An example of a compensating gait is shown in fig. S1B and movie S1.

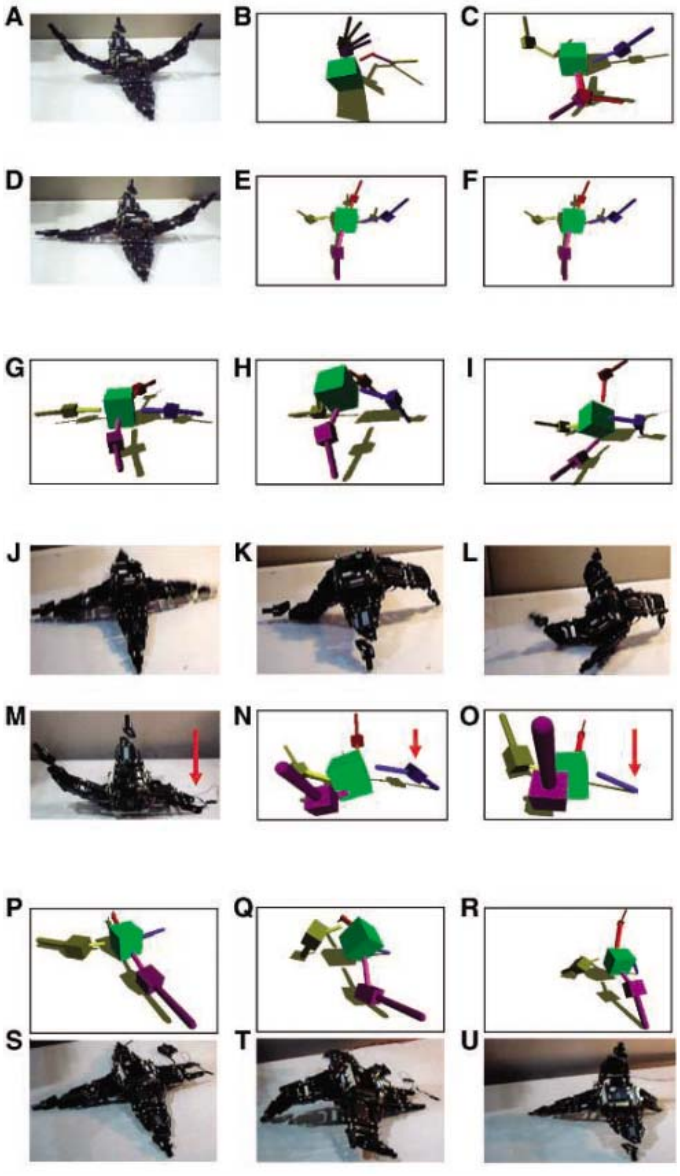

Fig. 2. The robot continually models and behaves. The robot performs a random action (**A**). A set of random models, such as (**B**), is synthesized into approximate models, such as (**C**). A new action is then synthesized to create maximal model disagreement and is performed by the physical robot (**D**), after which further modeling ensues. This cycle continues for a fixed period or until no further model improvement is possible (**E** and **F**). The best model is then used to synthesize a behavior. In this case, the behavior is forward locomotion, the first few movements of which are shown (**G** to **I**). This behavior is then executed by the physical robot (**J** to **L**). Next, the robot suffers damage [the lower part of the right leg breaks off (**M**)]. Modeling recommences with the best model so far (**N**), and using the same process of modeling and experimentation, eventually discovers the damage (**O**). The new model is used to synthesize a new behavior (**P** to **R**), which is executed by the physical robot (**S** to **U**), allowing it to recover functionality despite the unanticipated change.

Although the possibility of autonomous self-modeling has been suggested (*26*), we demonstrated for the first time a physical system able to autonomously recover its own topology with little prior knowledge, as well as optimize the parameters of those resulting self-models after unexpected morphological change. These processes demonstrate both topological and parametric self-modeling. This suggests that future machines may be able to continually detect changes in their own morphology (e.g., after damage has occurred or when grasping a new tool) or the environment (when the robot enters an unknown or changed environment) and use the inferred models to generate compensatory behavior. Beyond robotics, the ability to actively generate and test hypotheses can lead to general nonlinear and topological system identification (*23*) in other domains, such as computational systems (*22*), biological networks (*23*), damaged structures (*24*), and even automated science (*27*).

This work may inform future investigations of cognition in animals and the development of cognition in machines. Whereas simple yet robust behaviors can be created for robots without recourse to a model (*14*–*17*, *28*), higher animals require predictive forward models to function, given that in many cases biological sensors are

Table 1. Performance summary for the three algorithm variants. Baseline algorithms 1 and 2 disable the iterative and the model-driven nature of the learning process, respectively, while ensuring that the same computational effort and number of physical actions are used. Before damage, a successful experiment is determined as one that outputs a model with correct topology (see fig. S2 for examples of correct and incorrect topologies). Mean model error was calculated over the best model from each of the 30 experiments. Mean values are reported ± SD. An additional 90 experiments were conducted after the robot was damaged. The robot reinitiates modeling at this point using the most accurate model from the first 90 experiments (Fig. 2F). In this case, mean model error is determined as the difference between the inferred length of the damaged leg and the true damaged length (9.7 cm).

	Baseline 1	Baseline 2	Model-driven algorithm
Before damage			
Independent experiments (*n*)	30	30	30
Physical actions per experiment	16	16	16
Mean model evaluations (*n* = 30)	262,080 ± 13,859	246,893 ± 17,469	262,024 ± 13,851
Successful self-models	7	8	13
Success rate	23.3%	26.7%	43.3%
Mean model error (*n* = 30)	9.62 ± 1.47 cm	9.7 ± 1.45 cm	7.31 ± 1.22 cm
After damage			
Independent experiments (*n*)	30	30	30
Physical actions per experiment	16	16	16
Mean model evaluations (*n* = 30)	292,430 ± 44,375	278,140 ± 37,576	296,000 ± 22,351
Mean model error (*n* = 30)	5.60 ± 2.98 cm	4.55 ± 3.22 cm	2.17 ± 0.55 cm

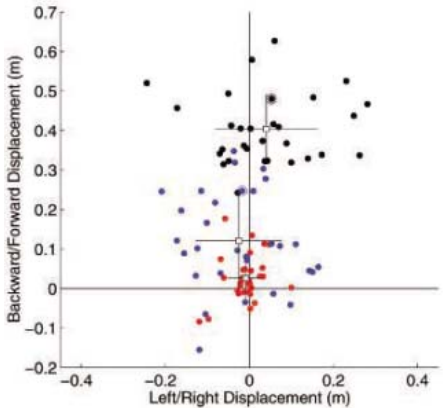

Fig. 3. Distance traveled during optimized versus random behaviors. Dots indicate the final location of the robot's center of mass, when it starts at the origin. Red dots indicate final positions of the physical robot when executing random behaviors. Black dots indicate final expected positions predicted by the 30 optimized behaviors when executed on the self-model (Fig. 2F). Blue dots denote the actual final positions of the physical robot after executing those same behaviors in reality. The behaviors corresponding to the circled dots are depicted in Fig. 2, G to L. Squares indicate mean final positions. Vertical and horizontal lines indicate 2 SD for vertical and horizontal displacements, respectively.

not fast enough to provide adequate feedback during rapid and complex motion (*29*). Although it is unlikely that organisms maintain explicit models such as those presented here, the proposed method may shed light on the unknown processes by which organisms actively create and update self-models in the brain, how and which sensor-motor signals are used to do this, what form these models take, and the utility of multiple competing models (*30*). In particular, this work suggests that directed exploration for acquisition of predictive self-models (*31*) may play a critical role in achieving higher levels of machine cognition.

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Supporting Online Material

www.sciencemag.org/cgi/content/full/314/5802/1118/DC1
Materials and Methods
Figs. S1 and S2
References
Movie S1

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Solid-State Thermal Rectifier

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We demonstrated nanoscale solid-state thermal rectification. High-thermal-conductivity carbon and boron nitride nanotubes were mass-loaded externally and inhomogeneously with heavy molecules. The resulting nanoscale system yields asymmetric axial thermal conductance with greater heat flow in the direction of decreasing mass density. The effect cannot be explained by ordinary perturbative wave theories, and instead we suggest that solitons may be responsible for the phenomenon. Considering the important role of electrical rectifiers (diodes) in electronics, thermal rectifiers have substantial implications for diverse thermal management problems, ranging from nanoscale calorimeters to microelectronic processors to macroscopic refrigerators and energy-saving buildings.

The invention of nonlinear solid-state devices, such as diodes and transistors, that control electrical conduction marked the emergence of modern electronics. It is apparent that counterpart devices for heat conduction, if they could be fabricated, would have

deep implications for thermal circuits, thermal management, and the field of phononics in general. In recent years, some theoretical proposals for thermal rectifiers have been put forward (*1*–*4*), but these usually require complex coupling between individual atoms and substrates that are difficult to achieve experimentally. However, as noted by Peierls (*5*), heat transport in one dimension can be anomalous, and the breakdown of Fourier's law in one-dimensional (1D) systems may be coupled with extraordinary nonlinear thermal effects (*6*), including rectification.

Nanotubes are nearly 1D and thus are ideal materials for exploring thermal rectification effects. Previous studies have demonstrated that the thermal conductivity of 1D carbon nanotubes (CNTs) and boron nitride nanotubes (BNNTs) is high and dominated by phonons (*7*, *8*). For unmodified nanotubes with uniform mass distribution, the thermal conductance is symmetric (i.e., independent of the direction of axial heat flow). To investigate asymmetric thermal propagation in a suitable 1D inhomogeneous medium, we modified CNTs and BNNTs so that they assumed a non-uniform axial mass distribution (Fig. 1).

Pristine multiwalled BNNTs were first synthesized by means of an adaptation of a previously reported method (*9*), yielding samples with a typical outer diameter of ~30 to 40 nm and a length of ~10 μm. High-quality CNTs with diameters ranging from 10 to 33 nm were prepared by means of conventional arc methods (*10*). Individual tubes were placed on a custom-designed microscale thermal conductivity test fixture (*11*), with the use of a piezo-driven manipulator operated inside a scanning electron microscope (SEM). In brief, the fixture incorporates independently suspended SiN_x pads, with symmetrically fabricated Pt film resistors serving as either heaters or sensors. One end of the nanotube was bonded to the heater, the other end was bonded to the sensor, and the body of the nanotube was suspended in the vacuum in between.

Figure 2A shows an SEM image of a multiwalled CNT mounted to the test fixture and B the corresponding low-magnification transmission electron microscopic (TEM) image of the same CNT. For thermal conductance measurements, a known power P was supplied to the heater while resistance changes of the heater and sensor were used to determine the resulting temperature changes of the heater (ΔT_h) and sensor (ΔT_s) pads. The thermal conductance K of the nanotube was determined from ΔT_h and ΔT_s with the use of the relation

$$K = \frac{P}{\Delta T_h - \Delta T_s}\left(\frac{\Delta T_s}{\Delta T_h + \Delta T_s}\right) \quad (1)$$

Because of unavoidable non-uniformities in the construction of the test fixture itself, the system with the attached pristine nanotube was first calibrated to establish residual asymmetry by switching the roles of the heater and sensor. All the measurements were done at room temperature.

Nanotubes were engineered in situ while mounted to the test fixture in the SEM. Trimethyl-cyclopentadienyl platinum ($C_9H_{16}Pt$) was deposited non-uniformly along the length of the nanotube in an attempt to achieve the non-uniform mass-loading geometry depicted in Fig. 1. Figure 2C shows a TEM image of the same CNT as in Fig. 2, A and B, after mass loading. The deposited $C_9H_{16}Pt$ was found to be amorphous and tightly bound to the CNT. The sample mass near the right contact has clearly been increased (Fig. 2C). Indeed, the mass loading is even more effective than Fig. 2C might suggest: The molecular weight of $C_9H_{16}Pt$ (~319 g/mol) is much larger than that of $(C–C)_5$ or $(BN)_5$ (~120 g/mol), and because the molecular volumes are similar, the mass density is correspondingly higher as well.

Depositing $C_9H_{16}Pt$ on a nanotube has several possible effects on the sample thermal conductance. The most obvious is that the fused $C_9H_{16}Pt$ forms an additional thermal conductance channel on parts of the sample. To test for the magnitude of this symmetrical enhancement, we suspended the fused $C_9H_{16}Pt$ across the test fixture pads and measured its thermal conductance. At room temperature, the thermal conductivity of $C_9H_{16}Pt$ was found to be temperature-independent and less than 1% of that of the nanotube. Hence, its direct thermal contribution can be neglected.

After mass loading, the thermal conductance of the nanotube was again tested in both directions. Thermal rectification of the nanotube is defined as

$$Rectification = \frac{K_{H\to L} - K_{L\to H}}{K_{L\to H}} \times 100\% \quad (2)$$

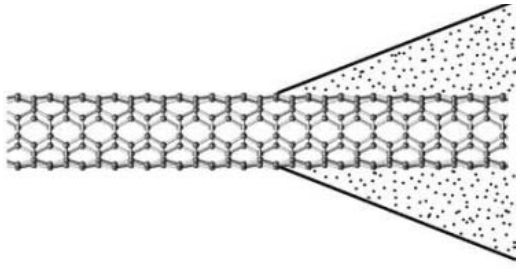

Fig. 1. A schematic description of depositing amorphous $C_9H_{16}Pt$ (black dots) on a nanotube (lattice structure).

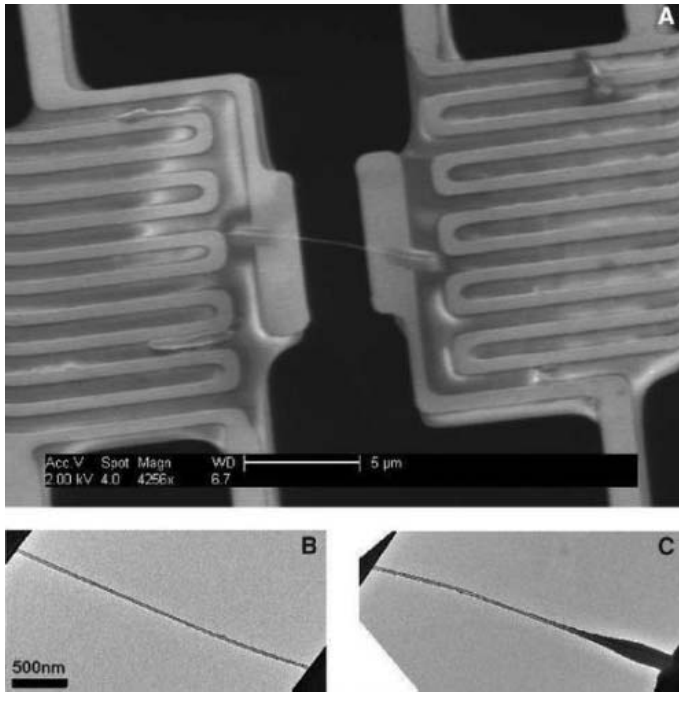

Fig. 2. (**A**) The SEM image of a CNT (light gray line in center) connected to the electrodes. Scale bar, 5 μm. (**B** and **C**) The corresponding low-magnification TEM images of the same CNT in (A), before (B) and after (C) $C_9H_{16}Pt$ was deposited.

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where $K_{L\rightarrow H}$ and $K_{H\rightarrow L}$ are the thermal conductances of the nanotube when heat flows from low-mass to high-mass ends or from high-mass to low-mass ends, respectively. For the CNT in Fig. 2, the measured thermal conductivity was 305 W/(m·K), and the rectification effect at room temperature was 2%.

Figure 3, A to C, shows three BNNTs that were also mass-engineered with $C_9H_{16}Pt$. The respective thermal rectifications were found to be 7, 4, and 3%. The arrows in Fig. 3 denote the direction of heat flow in which a higher thermal conductance was observed. All measurements showed that a higher thermal conductance was observed when heat flowed from the high-mass region (where more $C_9H_{16}Pt$ was deposited) to the low-mass region. Because electrons do not contribute to the thermal transport for BNNTs, the observed rectification effects can be attributed to nonelectronic excitations.

Figure 3D shows, in detail, the relation of ΔT_h versus ΔT_s for the BNNT of Fig. 3A before and after the deposition of $C_9H_{16}Pt$. Equation 1 can be expressed as $Ps/\Delta T_h(1-s^2)$, which, for $s \equiv \Delta T_s/\Delta T_h << 1$, reduces to $Ps/\Delta T_h$. Thus, the slope of the ΔT_h versus ΔT_s curve is proportional to absolute thermal conductance. $K_{L\rightarrow H}$ and $K_{H\rightarrow L}$ of the pristine nanotube are symmetric. After mass loading, $K_{H\rightarrow L}$ and $K_{L\rightarrow H}$ differ by 7%, well above the measurement uncertainty (~1%).

We now examine the origin of the observed thermal rectification. An asymmetric geometrical shape can, in principle, introduce asymmetric boundary scattering of phonons, whereby the thermal conductance can be reduced in one direction while it is increased in the other direction. In this scenario, thermal conductance is higher when heat flows from the narrow region to a wide region. Using the definition of Eq. 2, this always leads to a negative rectification coefficient, even for models where the boundary-scattering coefficient is mass-dependent. However, the thermal rectification observed in our experiments was always positive, and therefore any effect due to asymmetric shape was not dominant. Indeed, the sp^2 bonds in nanotubes are qualitatively much stronger than the bonds between fused $C_9H_{16}Pt$ molecules; thus, phonons should be mainly confined within the nanotubes, with relatively minor geometrical boundary-scattering effects.

A worthwhile analogy can also be made to photon wave propagation. The reflectivity R and the reflection coefficient r of a wave propagating across different media follow

$$R = r^2 = \left(\frac{k_i - k_t}{k_i + k_t}\right)^2 \qquad (3)$$

where k_i and k_t are the wave numbers of incident waves and transmitted waves, respectively. The squaring of the expression in Eq. 3 ensures that R is independent of the direction of incident waves. Because phonons are quanta of waves, the above result demonstrates that thermal rectification is not expected for ordinary wave transport. Similarly, impedance mismatching due to contact resistance will not lead to thermal rectification. In addition, nonlinear perturbative effects such as umklapp processes only decrease the total thermal conductance of the nanotube, without rectification.

Theoretical work has suggested the presence of stable solitons in nanotubes (*12*, *13*). Solitons are nonperturbative solutions of nonlinear systems. They are localized particle-like entities that can collide with each other without changing shape. Within a general class of soliton models, asymmetry of heat flow for an inhomogeneous medium is a common feature (*14*–*16*). As an example, the reflection amplitude r for the Korteweg–de Veries equation is (*14*)

$$r = \begin{cases} 0 & \left(v = \sqrt{\frac{m_2}{m_1}} \le 1\right) \text{ no soliton} \\ \left(\sqrt{2\frac{v-1}{v+1} + \frac{1}{4}} - \frac{1}{2}\right)^2 & \left(v = \sqrt{\frac{m_2}{m_1}} > 1\right) \text{ one soliton} \end{cases} \qquad (4)$$

where m_1 and m_2 are the mass of atoms whose displacement constitutes the incident and transmitted waves, respectively. The most important result of Eq. 4 is the asymmetry with respect to m_2/m_1. The direction of the thermal rectification is positive (better heat flow from high- to low-mass regions), which is consistent with the engineered nanotube rectification results presented above. For a crude estimate of the magnitude of the rectification effect, Eq. 4 yields a rectification of ~7% for a m_2/m_1 of ~5 [close to the molecular weight ratio of $C_9H_{16}Pt$ to $(C–C)_5$ or $(BN)_5$], consistent with the 2 to 7% rectification effects observed for mass-loaded nanotubes. Obviously, more-refined models of soliton transport in mass-loaded nanotubes, taking into account details of geometry, elastic constants, and mass distributions, are needed; but the key point is that linear or nonlinear perturbative systems do not lead to thermal rectification, whereas nonperturbative soliton models naturally do. The stronger ionic nature of BNNTs over CNTs also favors the nonlinearity. This may be the reason why BNNTs show a larger thermal rectification effect than CNTs.

With the availability of nonlinear thermal control, phonons should no longer be considered the unwanted by-products of electronics. Phonons, like electrons and photons, are information carriers and should be processed accordingly. Historically, semiconductor- or superconductor-based devices have been used to access thermal signals as soon as they are generated. Thermal rectifiers should make it possible to process thermal currents independently and convert them into electronic signals only when it is most efficient to do so.

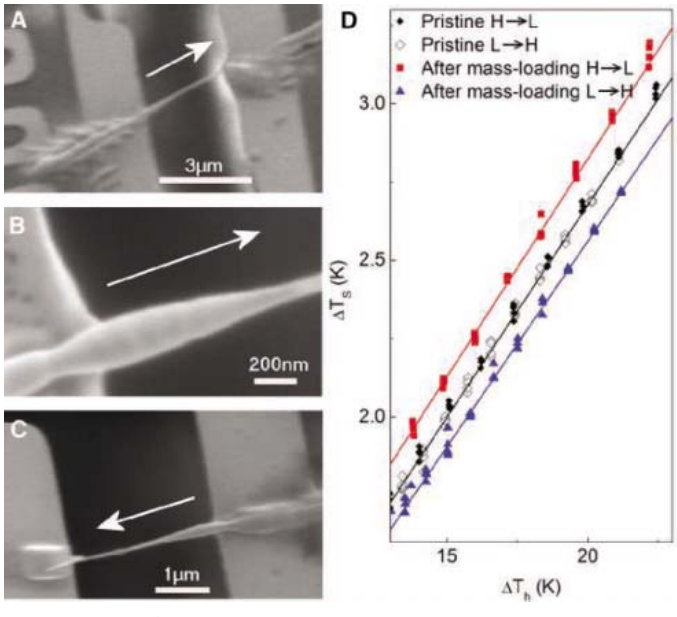

Fig. 3. (**A** to **C**) SEM images of three different BNNTs after deposition of $C_9H_{16}Pt$. The rectification measured was 7, 4, and 3%. The arrows denote the direction of heat flow, indicating where the thermal conductance is higher than that of the opposite direction. (**D**) Graphical representation of ΔT_h and ΔT_s for the BNNT in (A) before and after deposition of $C_9H_{16}Pt$. The solid lines are best-fit slopes intersecting the origin. For clarity, only data collected over a limited range of ΔT_h and ΔT_s are shown; data of similar quality were obtained over a much wider range of ΔT_h and ΔT_s.

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Reversible, Metal-Free Hydrogen Activation

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Although reversible covalent activation of molecular hydrogen (H_2) is a common reaction at transition metal centers, it has proven elusive in compounds of the lighter elements. We report that the compound $(C_6H_2Me_3)_2PH(C_6F_4)BH(C_6F_5)_2$ (Me, methyl), which we derived through an unusual reaction involving dimesitylphosphine substitution at a para carbon of tris(pentafluorophenyl) borane, cleanly loses H_2 at temperatures above 100°C. Preliminary kinetic studies reveal this process to be first order. Remarkably, the dehydrogenated product $(C_6H_2Me_3)_2P(C_6F_4)B(C_6F_5)_2$ is stable and reacts with 1 atmosphere of H_2 at 25°C to reform the starting complex. Deuteration studies were also carried out to probe the mechanism.

The generation and use of H_2 are important processes to fundamental chemical transformations (*1*–*7*) and biological functions (*8*). The overwhelming majority of systems known to either liberate or react with H_2 involve reaction at a transition metal center. Hydrogenase enzymes, as well as a plethora of synthetic stoichiometric and catalytic reagents for hydrogenation reactions, are based on the processes of oxidative addition and reductive elimination of H_2 at a metal center. Metal-free systems that either react with or liberate H_2 are rare. A unique metal-free hydrogenase from methanogenic archaea has been shown to catalyze reactions with H_2 (*9*–*11*), and theoretical studies suggest the role of a folate-like cofactor in the reversible activation or liberation of H_2 (*12*, *13*). Several metal-free systems have been shown to activate H_2. For example, main group element–H_2 reactions (*14*) in low-temperature matrices have been reported (*15*–*17*), and computational studies have probed the occurrence of H_2 bonds in main-group compounds (*18*, *19*). More recently, Power and co-workers (*20*) reported that the addition of H_2 to Ge_2-alkyne analogs affords a mixture of Ge_2 and primary germane products. Metal-free systems that liberate H_2 are of interest for their potential in H_2 storage applications. Although much effort has focused on hydride salts (*21*–*23*), a recent report by Thorn and co-workers describes an organic "hydride" system that reacts with protic compounds to eliminate H_2, although the assistance of a metal-based catalyst is required (*24*). Despite these advances, no metal-free system is yet reported to effect both the clean liberation and addition of H_2.

Here we report a phosphonium-borate species that undergoes thermally induced loss of H_2 to generate the corresponding phosphine-borane. We discovered this reaction sequence in the course of our studies on phosphine-borane interactions. The well-known Lewis acidic polymerization cocatalyst $B(C_6F_5)_3$ behaves as a traditional Lewis acid with donor molecules to form simple Lewis acid-base adducts (*25*, *26*). However, we have discovered that the sterically demanding secondary phosphine $(C_6H_2Me_3)_2PH$ reacts with $B(C_6F_5)_3$ to effect para-nucleophilic aromatic substitution, affording the zwitterionic phosphonium-borate $(C_6H_2Me_3)_2PH(C_6F_4)BF(C_6F_5)_2$ **1** (*27*) (Fig. 1).

The white, air- and moisture-stable solid **1** was isolated in 78% yield and exhibited a single phosphonium resonance in the $^{31}P\{^1H\}$ nuclear magnetic resonance (NMR) spectrum at –37.7 ppm as well as resonances in the ^{19}F NMR spectrum consistent with the presence of a BF bond and C_6F_4 and C_6F_5 rings. The corresponding ^{11}B NMR resonance revealed B-F coupling, and the ^1H NMR spectrum showed a doublet at 8.52 ppm from the PH fragment. Upon cooling to –15°C, the ^{19}F NMR resonances of the C_6F_4 bridge at –134 and –129 ppm split into doublets, consistent with inhibited rotation about the P–C_6F_4 bond. The thermodynamic barrier to this rotation was determined by variable-temperature NMR spectroscopy to be

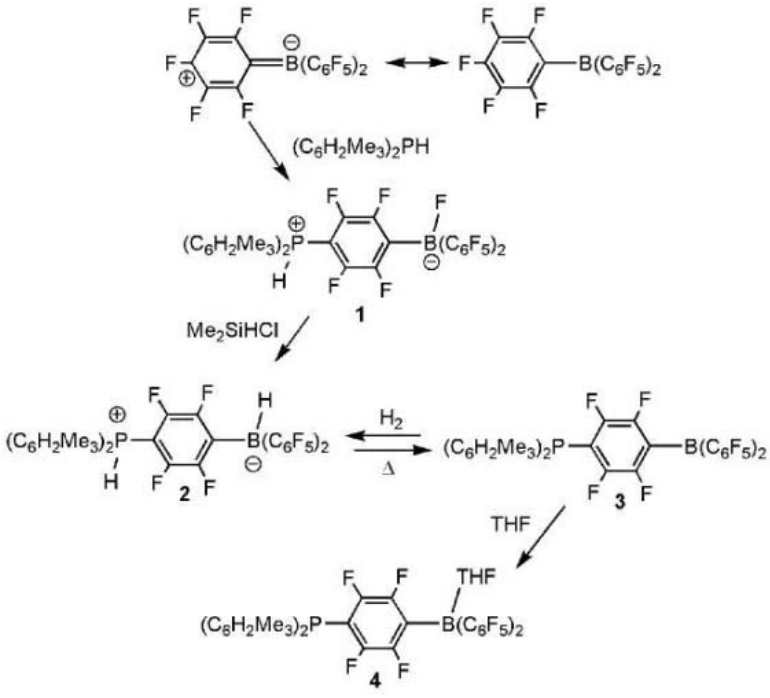

Fig. 1. Syntheses of compounds **1** to **4**.

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ΔG = 51.1 ± 0.5 kJ mol^{-1}. The formation of **1** implies that $(C_6H_2Me_3)_2PH$ is too large to coordinate to the B of $B(C_6F_5)_3$, prompting the observed aromatic substitution. In a similar fashion, Erker and co-workers reported the thermally induced rearrangement of an ylide-borane adduct $(Ph_3PCHPh)B(C_6F_5)_3$ (Ph, phenyl) to the para-substituted phosphonium-borate $(Ph_3PCHPh)(C_6F_4)BF(C_6F_5)_2$ (*28*).

Compound **1** rapidly reacted with Me_2SiHCl to give $(C_6H_2Me_3)_2PH(C_6F_4)BH(C_6F_5)_2$ **2** via H-for-F exchange. The white solid **2** exhibited a ^{11}B NMR signal at –25.2 ppm indicative of a four-coordinate boron hydride, whereas the $^{31}P\{^1H\}$ NMR resonance at –34.3 ppm was largely unchanged from that of **1**. The ^{1}H NMR spectrum showed doublet and quartet resonances at 8.49 ppm and 3.65 ppm, respectively, arising from the P-H and B-H fragments. A crystallographic study of **2** confirmed the proposed connectivity of $(C_6H_2Me_3)_2PH(C_6F_4)BH(C_6F_5)_2$ (*27*, *29*) (Fig. 2A). Thus, the air- and moisture-stable phosphonium-borate **2** can be prepared in a unique and facile two-step synthesis from readily available precursors (*27*).

In toluene solution, compound **2** underwent stoichiometric loss of H_2 in a facile manner upon heating above 100°C. The loss of H_2 was confirmed by the subsequent quantitative formation of the red-orange phosphinoborane species $(C_6H_2Me_3)_2P(C_6F_4)B(C_6F_5)_2$ **3** (*27*) (Fig. 1). It is noteworthy that phosphine-borane adducts of the form $R_2PH(BH_3)$ are also known to thermally or catalytically eliminate H_2 to give cyclic and polymeric phosphinoboranes (*30*, *31*). Monitoring the thermal decomposition of **2** by ^{19}F NMR spectroscopy showed a shift in the resonances attributed to the para-F of the C_6F_5 rings from –164 to –143 ppm, consistent with a change from four- to three-coordinate boron (*32*–*34*). Loss of the PH and BH resonances in the ^{1}H NMR and an upfield shift of 4 ppm in the $^{31}P\{^1H\}$ NMR spectrum confirmed the loss of H_2 from **2** and formation of **3**. Variable-temperature $^{31}P\{^1H\}$ NMR spectra of **3** revealed a barrier to P–C_6F_4 bond rotation of 42.7 kJ mol^{-1}, which is lower than that seen in **1**, consistent with the absence of H···ortho-F interactions in **3**. Weak π-donation from P and electron acceptance by B has been proposed for the related acetylene-based phosphinoborane $Ph_2PCCB(C_6H_2Me_3)_2$ (*35*, *36*); thus, on the basis of the intense red-orange color of **3** in solution (wavelength for maximum absorption λ-max, 455 nm; molar absorption coefficient ε = 487 liters cm^{-1} mol^{-1}) (Fig. 2C), it is tempting to attribute this color to an internal charge transfer.

The $^{31}P\{^1H\}$ NMR chemical shift for **3** showed minimal change with temperature, an observation consistent with the persistence of a pyramidal geometry at P. Nonetheless, polarization of charge in this donor-acceptor molecule may account for the observed color. Coordination of Lewis bases to B rendered the species colorless. As an example, recrystallization of **3** in the donor solvent tetrahydrofuran (THF) afforded colorless single crystals of the THF adduct **4**. This species exhibited NMR spectral data similar to **3** with additional resonances attributed to coordinated THF. A crystallographic study of **4** confirmed the formulation as $(C_6H_2Me_3)_2P(C_6F_4)B(THF)(C_6F_5)_2$ (*27*, *29*) (Fig. 2B); the geometry about B was pseudo-tetrahedral with THF coordination.

Remarkably, the isolated compound **3** reacted with H_2 in solution at 25°C. This reaction proceeded smoothly with rapid loss of the orange color to give a colorless solution of **2** (Fig. 2C). NMR data showed that the conversion to **2** was quantitative in less than 5 min. Thus, the thermally induced loss of H_2 from **2** was readily reversed. This reaction of **3** with H_2 was subsequently shown to be rapid even at –25°C. In a similar fashion, **3** reacted with D_2 to give the corre-

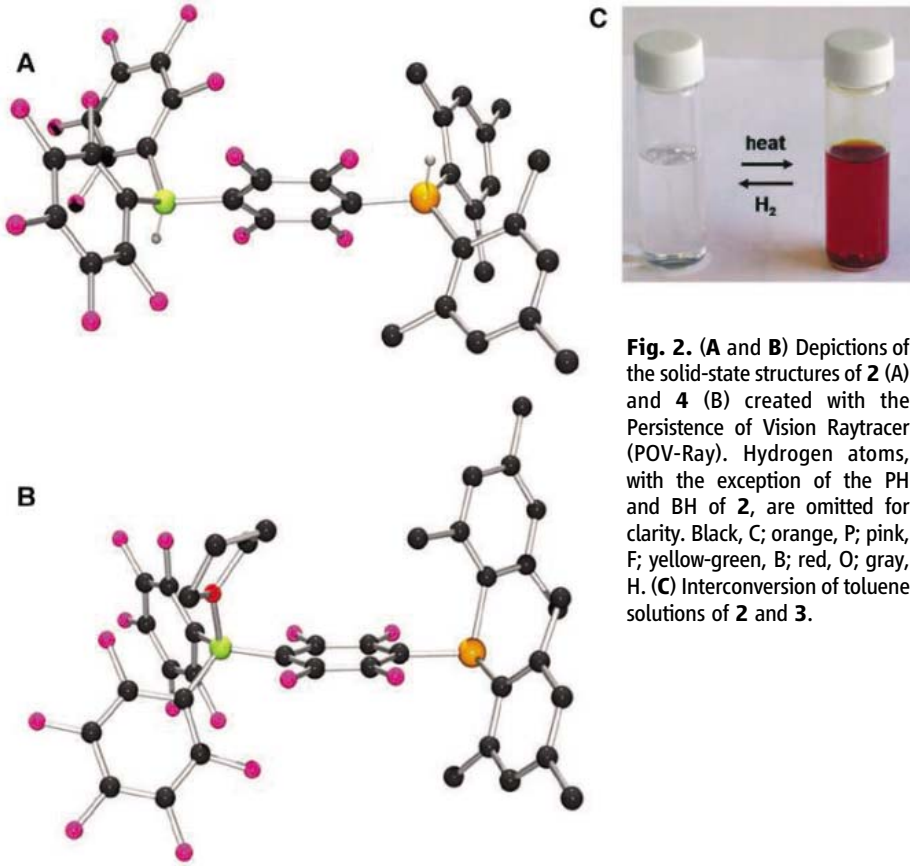

Fig. 2. (**A** and **B**) Depictions of the solid-state structures of **2** (A) and **4** (B) created with the Persistence of Vision Raytracer (POV-Ray). Hydrogen atoms, with the exception of the PH and BH of **2**, are omitted for clarity. Black, C; orange, P; pink, F; yellow-green, B; red, O; gray, H. (**C**) Interconversion of toluene solutions of **2** and **3**.

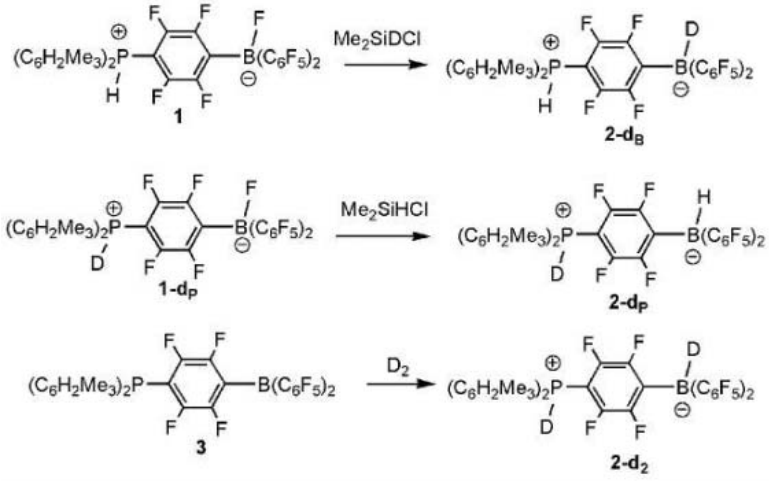

Fig. 3. Syntheses of the isotopomers of **2**.

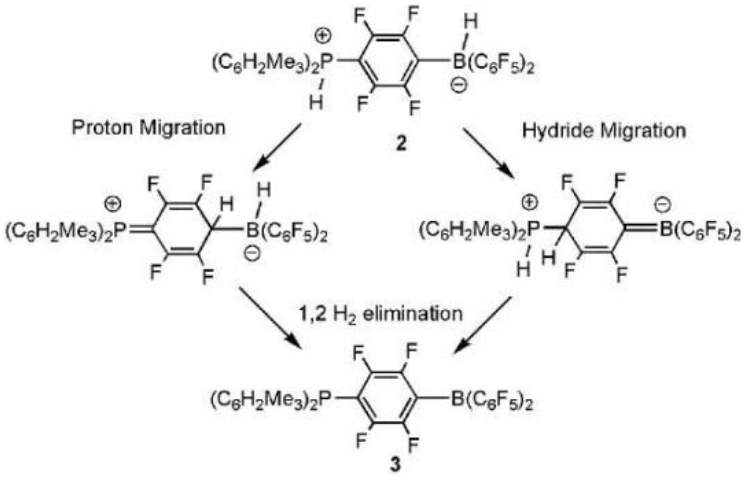

Fig. 4. Possible mechanisms for the formation of **3**.

sponding species $(C_6H_2Me_3)_2PD(C_6F_4)BD(C_6F_5)_2$ **2-d$_2$**. The site-specific labeled compound $(C_6H_2Me_3)_2PH(C_6F_4)BD(C_6F_5)_2$ **2-d$_B$** was prepared via reaction of **2** with Me_2SiDCl. Alternatively, the species $(C_6H_2Me_3)_2PD(C_6F_4)BH(C_6F_5)_2$ **2-d$_P$** was prepared following the procedure for **2** but using $(C_6H_2Me_3)_2PD$ to prepare **1-d$_P$**. These monodeuterated products (Fig. 3) showed no evidence of H-D exchange at 25°C; however, heating solutions of **2-d$_B$** or **2-d$_P$** to temperatures above 100°C in a sealed NMR tube, followed by cooling to 25°C, resulted in a statistical mixture of **2**, **2-d$_2$**, **2-d$_P$**, and **2-d$_B$**. This scrambling of H and D labels suggests the possibility of a high-temperature exchange process. To probe this finding further, we performed a conversion of a 1:1 mixture of **2** and **2-d$_2$** to **3**. The observation of HD in the statistical product mixture of H_2, D_2, and HD suggests a bimolecular high-temperature exchange process involving the intermolecular approach of BH and PH fragments in a transition state.

To gain further insight into this system, we acquired preliminary kinetic data. Initial rate data were obtained using $^{31}P\{^1H\}$ NMR spectroscopy to monitor the formation of **3** from the loss of H_2 by **2** in bromobenzene over the temperature range 100° to 150°C. Initially, spin-lattice relaxation time (T_1) measurements were performed to ensure that the relaxation delays were adequate to permit the correlation of integrals and concentrations. Over a concentration range of **2** from 0.02 M to 0.12 M, the consumption of **2** and generation of **3** were monitored over the first hour of reaction (*27*). These initial rate data showed that decay of the concentration of **2** followed first-order decay kinetics (figs. S1 and S2) with a rate constant of $3.5 \pm 0.6 \times 10^{-4}$ s^{-1} at 140°C. Eyring plots (fig. S3) over the temperature range 100° to 150°C provided the enthalpy of activation $\Delta H^{\neq} = 90 \pm 1$ kJ mol^{-1} and entropy of activation $\Delta S^{\neq} = -96 \pm 1$ J mol^{-1} K^{-1}. The entropy value and the first-order kinetics are consistent with an intramolecular process, and the enthalpy value suggests substantial bond breakage in the transition state. Intramolecular H_2 elimination requires proton and hydride on adjacent atoms. This could be achieved by proton migration from P to the C adjacent to B, or alternatively by hydride migration from B to the C adjacent to P (Fig. 4). The present data do not allow us to explicitly distinguish between these possibilities. We speculate, on the basis of considerations of the microscopically reverse reaction where the uptake of H_2 by **3** is intuitively thought to be initiated by the interaction of H_2 with B, that proton migration is more likely. This view is supported by the fact that **4** did not react with H_2.

This reaction system demonstrates that reversible small-molecule activation is achievable in the absence of a transition metal. This finding foreshadows new vistas in metal-free reactions and catalysis. Similarly, although the present system reversibly binds less than 0.25 weight % H_2, which is much less than the targets of 6 to 9%, it does suggest that new strategies for chemical hydrogen storage may involve Lewis acid–Lewis base cooperative reactivity.

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37. Supported by the Natural Sciences and Engineering Research Council (NSERC) of Canada, a NSERC graduate scholarship (G.C.W.), and an Ontario graduate scholarship (J.D.M.). We thank J. Stryker for helpful discussions. Structural parameters for compounds **2** and **4** are available free of charge from the Cambridge Crystallographic Data Centre under reference numbers CCDC-621908 and CCDC-296070.

Supporting Online Material

www.sciencemag.org/cgi/content/full/314/5802/1124/DC1
Materials and Methods
Figs. S1 to S3
Tables S1 and S2
References

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Bedrock Fracture by Ice Segregation in Cold Regions

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The volumetric expansion of freezing pore water is widely assumed to be a major cause of rock fracture in cold humid regions. Data from experiments simulating natural freezing regimes indicate that bedrock fracture results instead from ice segregation. Fracture depth and timing are also numerically simulated by coupling heat and mass transfer with a fracture model. The depth and geometry of fractures match those in Arctic permafrost and ice-age weathering profiles. This agreement supports a conceptual model in which ice segregation in near-surface permafrost leads progressively to rock fracture and heave, whereas permafrost degradation leads episodically to melt of segregated ice and rock settlement.

The fracture of bedrock is fundamental to debris production and landscape development. Rock fracture in polar and alpine regions has often been attributed to the freezing and volumetric expansion of water trapped within pores and cracks (*1*). An alternative process of bedrock fracture, involving ice segregation, remains poorly characterized despite a number of theoretical and experimental studies over the past two decades (*2*–*8*). Ice segregation occurs when temperature gradient–induced suction in freezing or frozen ground drives unfrozen water—held in capillaries and adsorbed on the surfaces of mineral particles—through a porous medium, such as soil, toward freezing sites where lenses or layers of ice grow. If ice segregation fractures bedrock permafrost, the fractures and ice lenses are expected to concentrate in wet, porous rock just beneath the top of the permafrost and in the base of the active layer (*3*, *5*–*7*), as they do in porous silty soils (*9*, *10*). Here, we report results from experiments that test this hypothesis and elucidate the ice-segregation process in bedrock.

We developed an experimental methodology in a pilot study (*5*) that instrumented a block of chalk; the block's lower half was maintained at temperatures below 0°C (simulating permafrost) and its upper half cycled above and below 0°C (simulating seasonal thawing and freezing of the overlying active layer). We then performed systematic experiments with 10 substantially larger chalk blocks that had different moisture contents in order to simulate and monitor bidirectional freezing of a bedrock active layer above permafrost and unidirectional freezing of seasonally frozen bedrock (*11*). Bidirectional freezing refers to the combination of upward freezing from the permafrost table and downward freezing from the rock surface, whereas unidirectional freezing refers simply to the latter. The results validated those of the pilot study (*6*) and revealed discrete stages of micro- and macrocrack development. Based on numerical simulations of the depth and timing of rock fracture, we propose a conceptual model of ice segregation, fracture development, and surface stability in porous bedrock permafrost.

The experiments demonstrate that ice segregation fractures wet chalk. We discounted volumetric expansion as a cause of fracture because measured rock saturation levels at the depths of fracture in the bidirectional experiments did not exceed ~65% when macrofracture growth commenced, indicating that fracturing occurred at moisture levels substantially below the predicted threshold saturation level of ~91% (*12*). In addition, sustained periods of rock-surface heave during thawing cycles (Fig. 1) are inconsistent with bursts of crack growth predicted by volumetric expansion during freezing cycles, whereas they are expected with sustained periods of ice segregation (*12*). Thaw-related rock heave (Fig. 2) is analogous to summer frost heave in Arctic permafrost soils (*13*), when unfrozen water migrates down into underlying frozen ground, permitting ice-lens growth in still-frozen soil beneath the thawing front. Fractures formed by ice segregation should generally be parallel to the cooling surfaces (*14*), as we observed. All but the thinnest fractures in the artificial permafrost contained visible segregated ice (Fig. 3, A and C).

The freezing regime determines the depth of rock fracture. Bidirectional freezing results in fracture of the upper layer of permafrost and the base of the active layer, whereas unidirectional freezing results in fracture close to the surface (Fig. 3). This distinction reflects seasonal differences in the directions of heat and water transfer. Unidirectional (downward) freezing produces an upward transfer, favoring ice segregation near the rock surface. Bidirectional freezing causes transfer from the center of the active layer down toward the permafrost and up toward the rock surface. The concentration of segregated ice in the upper layer of permafrost and the base of the active layer results in part from downward transfers associated with the slower rate of upward freezing, allowing more time for water migration. But the major factor, as indicated by the seasonal timing of rock heave (Fig. 1), is summer ice segregation beneath the permafrost table.

Fractures within permafrost develop in two stages. We attributed gradual progressive heave during initial seasonal temperature cycles to microcrack development (Fig. 1). A threshold is then crossed, after which rapid heave commences during thawing cycles (summers), recording the rapid growth of segregated ice in macrocracks. During these subsequent seasonal temperature cycles, ice

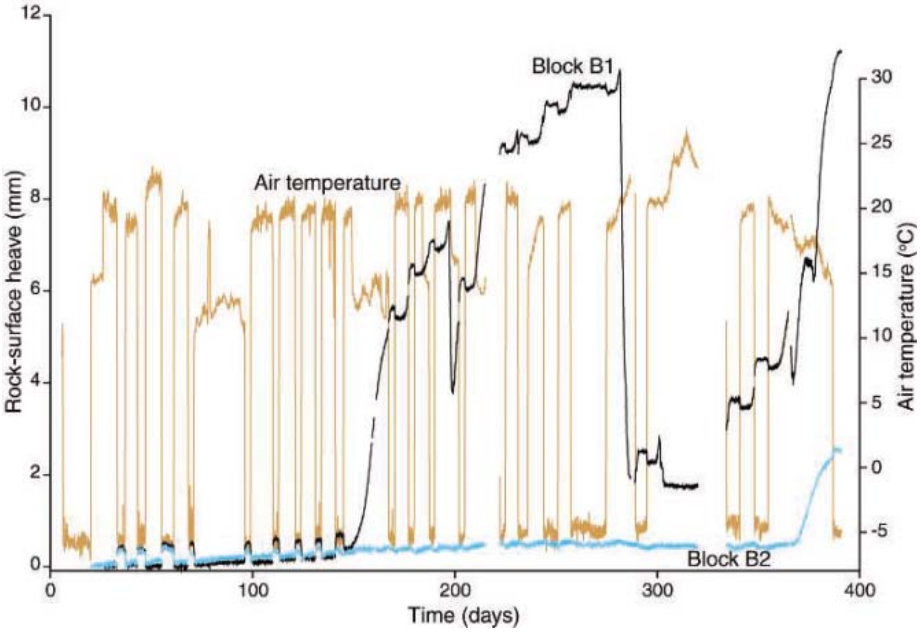

Fig. 1. Rock-surface heave and air temperature during 21 cycles of bidirectional freezing of blocks B1 and B2 (*11*). Air temperatures above 0°C correspond with thawing cycles, and those below 0°C correspond with freezing cycles. The data are 4-hour running means. Thresholds in the heave data at day 150 for block B1 and day 370 for block B2 mark the change from micro- to macrocracking, and the onset of development of an ice-rich fractured layer of permafrost. Thaw settlement of nearly 10 mm at the surface of B1 (day 283) coincided with the European heat wave of July and August 2003.

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segregation forms an ice-rich layer of fractured rock just below the permafrost table (Fig. 3A). The icy layer is sensitively dependent on summer air temperatures, rapidly melting during the European heat wave of summer 2003 (~day 283 on Fig. 1), but re-forming afterward.

We performed numerical simulations of the experiments by coupling a description of transient heat and mass transfer in porous freezing media with the segregation ice–induced fracture model of Walder and Hallet (*2*). Assuming an initially uniform distribution of very small (5-mm) defects throughout the block (*11*), the simulations successfully predict the approximate depth of maximum cracking for both the unidirectional (Fig. 4A) and bidirectional (Fig. 4B) freezing experiments. Notably smaller cracks are predicted during unidirectional freezing. The simulations also provide insight into the interdependence of temperature, temperature gradient, and pressure within a growing microcrack, and the conditions necessary for eventual fracture. The three conditions necessary for crack growth due to ice segregation are a subzero (°C) temperature gradient, to cause a thermomolecular pressure gradient; a warm (slightly subzero) temperature, for sufficient water permeability; and an intracrack pressure moderately above the stress-corrosion limit (*2*). When all three conditions are met simultaneously, cracks that begin small can lengthen almost exponentially (Fig. 4C). During cyclic freeze-thaw events, the pressure within a crack also cycles, increasing when water is drawn inside by the thermomolecular pressure gradient and frozen, and decreasing when ice thaws and pressurized water is forced out (Fig. 4D). As a crack grows, the pressure required to lengthen it

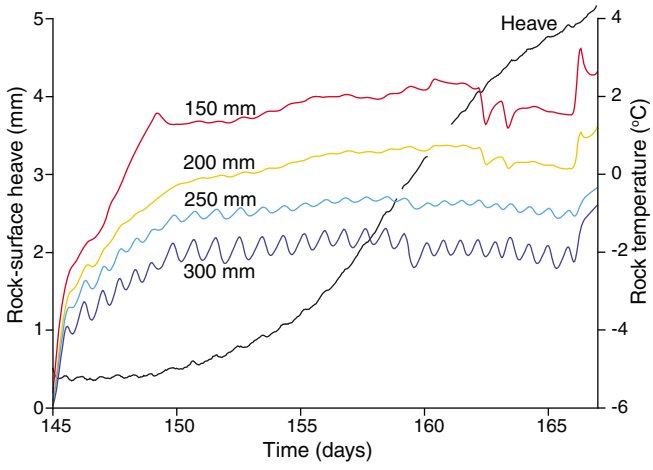

Fig. 2. Rock temperature in the lower part of the active layer (150- and 200-mm depths) and the upper part of the permafrost (250- and 300-mm depths), and rock-surface heave during thaw cycle 10, block B1. The active layer attained a depth of 241 mm on day 167.

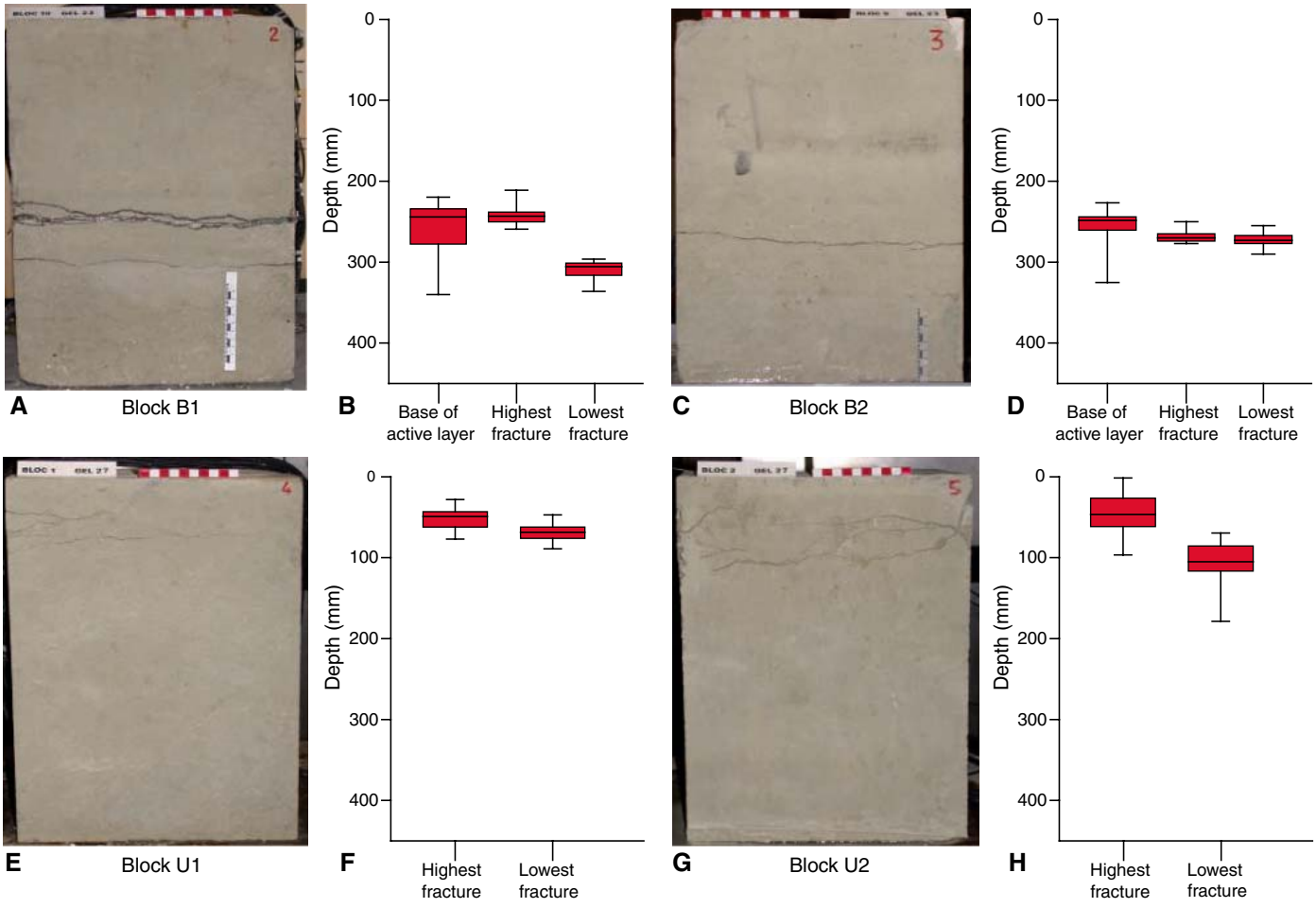

Fig. 3. Fractures and segregated ice within 450-mm-high blocks of chalk subjected to bidirectional freezing (**A** to **D**) and unidirectional freezing (**E** to **H**). The photographs show vertical sections through frozen blocks at the end of the experiments, with lenses or layers of segregated ice visible in many cracks. The box plots show fracture depths and, for the bidirectional experiments, the base of the active layer (the depth of maximum seasonal penetration of the 0°C isotherm into the rock). The red boxes show the interquartile range, the horizontal line within the boxes is the median, and the whiskers extend to the highest and lowest values. In the unidirectional experiments, the depth of maximum penetration of the 0°C isotherm exceeded 350 mm during each freezing cycle.

further decreases as a result of the increasing stress intensity factor (the same phenomenon causes small windshield cracks to grow quickly). The rapid increase in rock-surface heave around 150 days in Fig. 1 correlates well with the predicted time that a crack at 300-mm depth would reach 300 mm in length, effectively fracturing the block in two and allowing for relatively unrestrained ice accumulation within the fracture. Although the exact time for complete fracture depends on the initial defect size (5 mm), order-of-magnitude variations in initial size (0.5 to 50 mm) still lead to complete fracture within the 125- to 175-day range. The model results agree with the theory of Walder and Hallet (*2*) and also provide insight into the effects of a cyclic thermal regime (supporting online material text).

The layer of ice-rich fractured chalk near the top of the artificial permafrost (Fig. 3A) resembles ice-rich fractured limestones, shales, and sandstones in Arctic permafrost. The natural fractures have been attributed to both thermal contraction cracking (*15*) and ice segregation (*14*, *16*), and the ice has been variably interpreted as hoar frost (*15*) or segregated ice (*14*, *16*). We discounted thermal contraction cracking because, in frozen soils, it produces large widely spaced vertical cracks and ice wedges (*17*), whereas the majority of fractures in the artificial and natural permafrost are small, closely spaced, and horizontal, consistent with horizontal cooling surfaces during ice segregation. The experimental fracture patterns also resemble those in weathering profiles from regions where ice-age permafrost previously existed. Fractures of the upper ~0.5 to several meters of bedrock are abundant in fine-grained porous rocks such as the chalk in northern France (*18*) and southern England (*19*), the muschelkalk (shelly limestone) of central Germany (*15*), and various limestones, mudstones, and clays in the United Kingdom (*19*). In all cases, the fractures are mostly horizontal to subhorizontal, and they decrease in number density as depth increases. We could not discount the possibility, however, that some horizontal cracks result from other weathering processes or preexisting weaknesses, such as bedding planes.

The agreement between modeling and field observations supports the hypothesis that ice segregation preferentially fractures porous bedrock near the permafrost table and contributes substantially to a more precise understanding of the ice-segregation process in bedrock. The wider importance of this process to geomorphology, Quaternary science, engineering, and climate change concerns the dynamics of the ice-rich fractured layer in the upper meters of permafrost. Ice segregation leads progressively to rock fracture, accumulation of ground ice, and rock heave, whereas active-layer deepening leads episodically to melt of ground ice and rock settlement. The interplay between the buildup and decay of the ice-rich layer operates on a number of time scales. During unusually hot summers, the development of this layer is interrupted by thaw settlement (Fig. 1), just as in ice-rich permafrost soils (*20*), when the top of the permafrost thaws and supplies rock fragments to the active layer. The fragments are then available for sorting into patterned ground (*21*), comminution by weathering, or downslope transport by mass wasting, fluvial activity, or glaciers. During climate warming at the end of Quaternary glacial or stadial periods, the ice-rich layer in mid-latitude regions such as the chalklands of England and France melted completely, triggering thaw settlement of the ground surface, deformation of soil and rock, and enhanced mass wasting (*22*). With recent climate warming predicted to continue during the next century, amplified in Arctic and sub-Arctic regions (*23*), the warming and thawing of ice-rich bedrock permafrost is likely to destabilize many rock substrates. Such issues will undoubtedly become important to scientists, engineers, and inhabitants of permafrost regions.

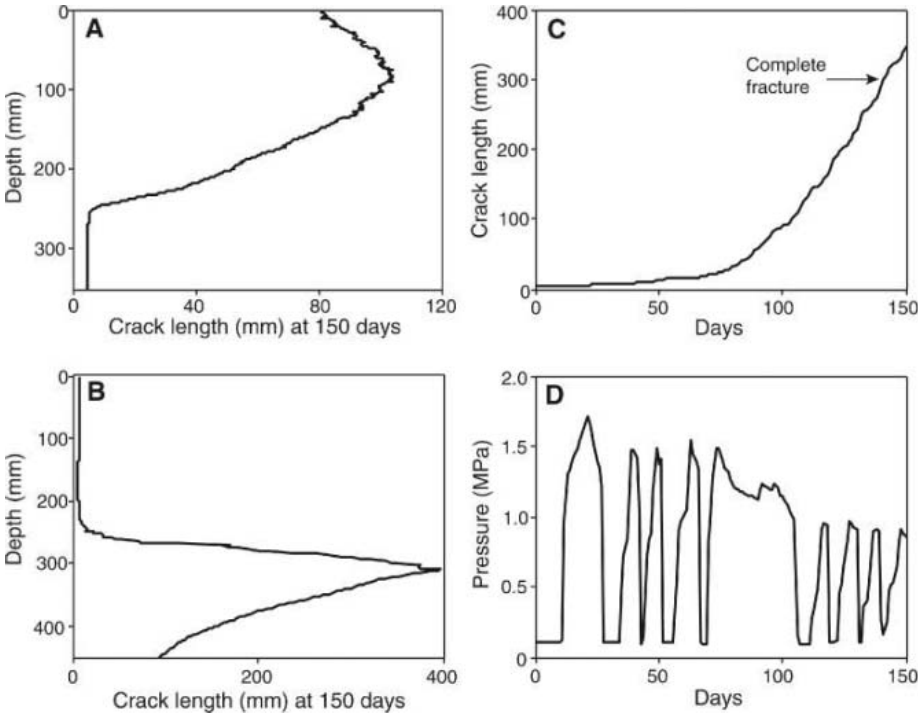

Fig. 4. Numerical model predictions showing the final crack length at 150 days as a function of vertical location of the initial crack for (**A**) unidirectional freezing and (**B**) bidirectional freezing. During bidirectional freezing, an initial crack at a depth of 300 mm begins to rapidly lengthen around day 75 (**C**). Complete fracture denotes the time when the crack length becomes equal to the block width (300 mm). The intracrack pressure during bidirectional freezing fluctuates between a maximum during ice accumulation and a minimum after thaw (**D**).

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Materials and Methods
SOM Text
Fig. S1
Table S1
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The Impact of Boreal Forest Fire on Climate Warming

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We report measurements and analysis of a boreal forest fire, integrating the effects of greenhouse gases, aerosols, black carbon deposition on snow and sea ice, and postfire changes in surface albedo. The net effect of all agents was to increase radiative forcing during the first year (34 ± 31 Watts per square meter of burned area), but to decrease radiative forcing when averaged over an 80-year fire cycle (−2.3 ± 2.2 Watts per square meter) because multidecadal increases in surface albedo had a larger impact than fire-emitted greenhouse gases. This result implies that future increases in boreal fire may not accelerate climate warming.

Arctic and boreal regions are warming rapidly, with multiple consequences for northern ecosystems and global climate (*1*). In boreal ecosystems, future increases in air temperature may lengthen the fire season and increase the probability of fires, leading some to hypothesize a positive feedback between warming, fire activity, carbon loss, and future climate change (*2*, *3*). Although CO_2 and other greenhouse gases emitted by fire contribute to climate warming, understanding the net effect of a changing fire regime on climate is challenging because of the multiple ways by which fires influence atmospheric composition and the land surface. Emissions of aerosols, for example, can lead to either warming or cooling at a regional scale, depending on factors such as aerosol composition and the underlying albedo of both the Earth's surface and clouds (*4*). Subsequent deposition of black carbon aerosols on glaciers, snow, sea ice, and the Greenland ice sheet may reduce surface albedo (*5*), causing both atmospheric heating (*6*) and enhanced surface melting. Within a burn perimeter, combined changes in ecosystem structure and species composition after fire cause net radiation and sensible heat fluxes to decline substantially (*7*, *8*). These changes in the local surface energy budget persist for decades and are probably regionally variable. Concurrently, accumulation of carbon in organic soils and vegetation during intermediate successional stages offsets the pulse of carbon released during combustion (*9*).

Understanding the net effect of these processes (and their temporal and spatial scales) is important in managing northern forests to mitigate the climate impacts of fossil fuel emissions. Although changes in boreal forest albedo can have a considerable cooling effect on Northern Hemisphere climate (*10*, *11*), these changes are offset by accompanying changes in carbon accumulation (*12*), so the net effect of land cover change on climate may be close to neutral at a global scale when both surface energy balance and CO_2 fluxes are considered (*13*). Here we applied the concept of radiative forcing (*12*) to assess quantitatively the net effect of a boreal forest fire on climate, on the basis of carbon and surface energy budget measurements that we made in a fire chronosequence of black spruce (*Picea mariana*) in interior Alaska. We considered two time scales: the year immediately after fire and an 80-year period during which species composition and ecosystem structure returned to a prefire mature successional state as defined by an adjacent unburned control stand.

The Donnelly Flats crown fire occurred during 11 to 18 June 1999 in interior Alaska (63°55′N; 145°44′W) and burned ~7600 ha (*14*). The fire was intense (e.g., figs. S1 and S2), causing stand-replacing mortality of the black spruce within the burn perimeter and consuming much of the soil organic matter above the mineral horizon (*15*). Aboveground fuel consumption from overstory and understory vegetation was estimated with a combination of harvesting, allometry, and inventory methods. Postfire soil respiration losses during the first year after fire were estimated with a combination of chamber measurements and eddy covariance measurements. Precision spectral pyranometers (Eppley Laboratory, Inc., Newport, RI) measured incoming and outgoing shortwave radiation above the canopy (and thus surface albedo) during July and August of 1999 within the burn perimeter (*7*) and then mostly continuously from October 1999 through September 2004 at both the burn and control.

We converted field measurements of carbon loss during the fire to CH_4 and CO_2 fluxes using emission factors (*16*). Radiative forcing from these greenhouse gases was estimated with equations derived from a global radiative transfer model (*17*). In our figures and table, we report global annual mean radiative forcing (in W) per m^2 of burned area, with radiative forcing defined following the Intergovernmental Panel on Climate Change Third Assessment Report convention as the change in net radiation at the tropopause after stratospheric ad-

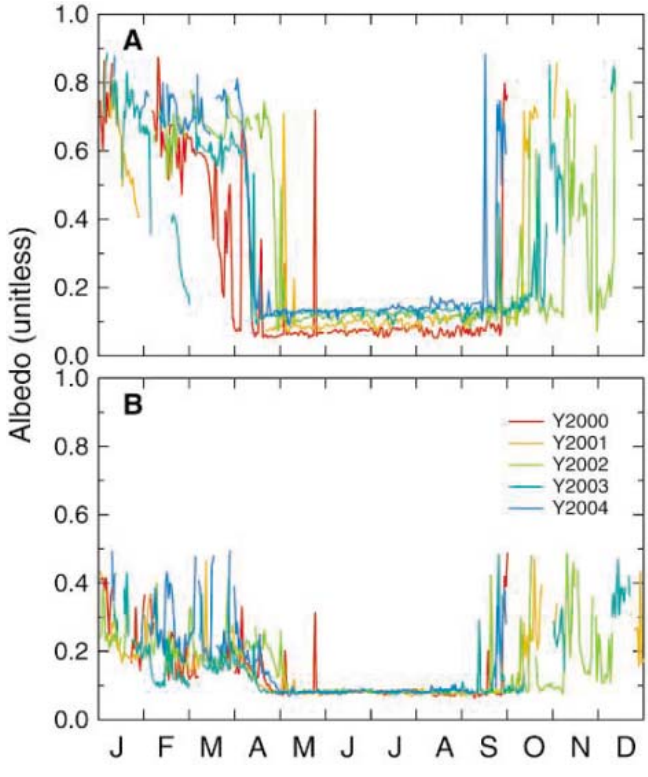

Fig. 1. Midday surface albedo within the burn perimeter of the Donnelly Flats fire (**A**) and from the adjacent black spruce stand that served as a control (**B**). Summer albedo progressively increased during each year and exceeded values at the control site ~3 years after fire. Snow events, including one in late May of 2000, caused spikes that are visible at both the burn and control sites.

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justment (*18*). CH_4 was assumed to have a 10-year atmospheric lifetime. The lifetime of the CO_2 anomaly from the fire was estimated with a combination of ocean impulse-response functions from the Joos and Siegenthaler ocean carbon model (*19*) and a postfire trajectory of net ecosystem production (NEP) that we constructed using mass balance constraints and eddy covariance measurements (figs. S3 and S4). We used the Column Radiation Model (*20*) to estimate radiative forcing from changes in surface albedo within the Donnelly Flats burn perimeter (figs. S5 to S8). The persistence of albedo changes in postfire ecosystems was assessed from an analysis of MODerate Resolution Imaging Spectroradiometer (MODIS) albedo measurements (*21*) within burn perimeters of known ages (*14*) across interior Alaska. We derived radiative forcing from the fire-induced ozone anomaly using simulations from the National Center for Atmospheric Research Community Atmosphere Model version 3 (CAM 3) of the 2004 Alaska and Yukon fire complex (*22*) scaled to the carbon emission levels that we measured for the Donnelly Flats fire. Similarly, we estimated radiative forcing from the direct effect of aerosols and deposition of black carbon on snow and sea ice by injecting emissions from the Donnelly Flats fire into CAM 3 (*23*, *24*). In the Supporting Online Material we provide more information about our methods for estimating radiative forcing, as well as an additional set of forcing estimates that take into account the efficacy of the different agents (*25*).

During the fire event, 206 ± 110 g C m^{-2} were emitted by combustion from the black spruce overstory, 107 ± 74 g C m^{-2} from the vascular plant understory, and 1246 ± 600 g C m^{-2} from the duff layer composed of mosses, lichens, roots, partially decomposed plant litter, and humus. Total fuel consumption for the Donnelly Flats fire (1560 ± 610 g C m^{-2}) was similar to other estimates for boreal North America, including 1580 g C m^{-2} for moderately severe fires in boreal North America (*26*) and 1300 g C m^{-2} for the mean of Canadian boreal forests (*27*). Including additional soil respiration losses of 202 ± 53 g C m^{-2} $year^{-1}$during the first year after fire, the ecosystem lost a total of 1760 ± 620 g C m^{-2}.

Radiative forcing from long-lived greenhouse gases (CH_4 and CO_2) contributed a total of 8 ± 3 W m^{-2} during the first year. Deposition of black carbon on snow and sea ice added another 8 ± 5 W m^{-2}. An increase in tropospheric ozone from fire-emitted trace gases generated a positive radiative forcing of 6 ± 4 W m^{-2}. Fire-emitted aerosols mixed widely across arctic and boreal regions (fig. S9), decreased net radiation at the surface (−90 ± 35 W m^{-2}), but did not substantially change radiative forcing (17 ± 30 W m^{-2}). Changes in surface albedo within the fire perimeter offset positive radiative forcing from the other agents. Specifically, the loss of overstory canopy after fire led to increased snow exposure during spring and fall (fig. S10), higher albedo (Fig. 1), and a negative annual radiative forcing (−5 ± 2 W m^{-2}). The combined effect of all forcing agents was 34 ± 31 W m^{-2} during year 1 (Table 1).

After the first year, the short-lived effects of ozone, aerosols, and black carbon deposition were no longer important, so the net effect of the fire on radiative forcing reflected the balance between the persistence of postfire changes in surface albedo and the effects from the remaining greenhouse gases in the atmosphere. During the first 5 years after fire, summer albedo progressively increased (Fig. 1), probably from an increase in grass and shrub cover and partial loss of black carbon that initially coated soil surfaces and dead black spruce boles. This strengthened the negative radiative forcing from postfire albedo changes, with this quantity decreasing from −5 ± 2 W m^{-2} during the first year to −8 ± 3 W m^{-2} during the period 3 to 5 years after fire. Analysis of MODIS satellite data from nearby forest stands provided evidence that spring and summer albedo typically remains elevated for about three decades after fire and that recovery to prefire albedo levels requires ~55 years (Fig. 2).

We predicted that the greenhouse gas pulse from the Donnelly Flats fire should gradually decline over a period of 5 decades, owing to CH_4 oxidation and CO_2 uptake by the oceans and regrowing vegetation within the burn perimeter (fig. S3D). During this interval, the greenhouse gases will contribute to a positive radiative forcing

Table 1. Radiative forcing associated with the Donnelly Flats fire.

Forcing agent	Radiative forcing* [W (m^2 burned)$^{-1}$]	
	Year 1	Years 0 to 80 (mean)
Long-lived greenhouse gases (CH_4 and CO_2)	8 ± 3	1.6 ± 0.8
Ozone	6 ± 4	0.1 ± 0.1
Black carbon deposition on snow	3 ± 3	0.0 ± 0.0
Black carbon deposition on sea ice	5 ± 4	0.1 ± 0.1
Aerosols (direct radiative forcing)†	17 ± 30	0.2 ± 0.4
Impact at the surface: −90 W ± 35 m^{-2}		
Changes in post-fire surface albedo	−5 ± 2	−4.2 ± 2.0
Total‡	34 ± 31	−2.3 ± 2.2

*All the radiative forcing estimates reported here represent annual mean values (in W) for the global atmosphere associated with burning of a 1-m^2 area within the perimeter of the Donnelly Flats fire. We report values averaged over year 1 and for the mean of the 0- to 80-year period after fire (and including the fire event). †We did not estimate indirect effects of aerosols on radiative forcing as mediated, for example, by cloud drop sizes or cloud lifetime (*4*). Although uncertain, indirect aerosol effects are thought to contribute to negative radiative forcing (*18*, *25*) and would offset other positive radiative forcing agents during year 1. ‡Accounting for the efficacy of the different forcing agents (*25*), the total effective forcing of the Donnelly Flats fire was 18 ± 42 W m^{-2} during year 1 and −2.4 ± 2.3 W m^{-2} during years 0 to 80 (tables S1 and S2).

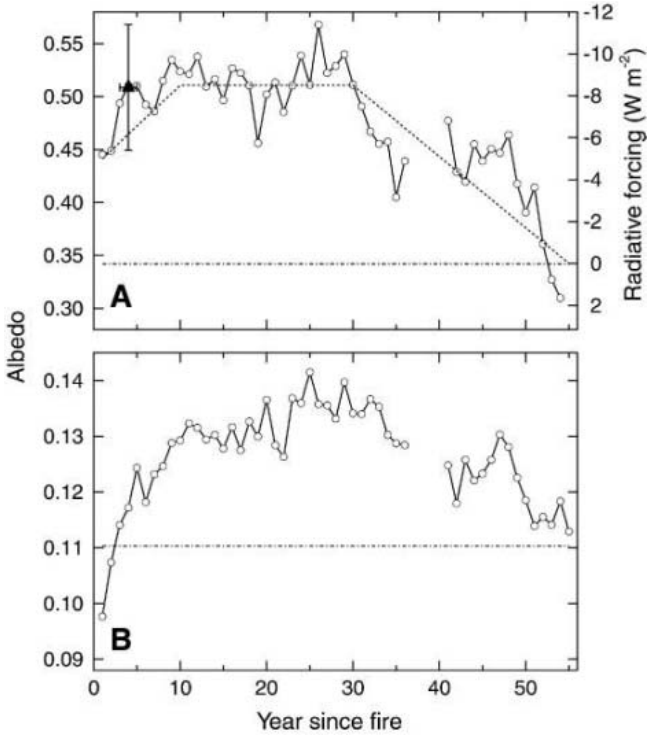

Fig. 2. Postfire albedo during (**A**) spring (Julian Days 33 to 113) and (**B**) summer (Julian Days 145 to 241) from MODIS satellite observations extracted from burn scars of different ages in interior Alaska (circles and solid line, left axis). A control was constructed from the mean of evergreen conifer vegetation that did not burn in the last 55 years (dashed-dotted line, left axis). Annual radiative forcing as estimated from tower measurements of albedo from burn and control sites during 2002 to 2004 was −8 ± 3 W m^{-2} [(A), triangle, right axis]. The longer-term postfire trajectory of albedo-driven annual radiative forcing was assumed to follow the MODIS albedo pattern [(A), dashed line, right axis]. Years with limited burned area were excluded from the analysis.

(Fig. 3A). After ~60 years, continued uptake by the postfire ecosystem should cause atmospheric CO_2 to decrease below background levels, subsequent withdrawal of CO_2 from the ocean, and a negative radiative forcing. As a result of this trajectory and concurrent changes in surface albedo, the influence of the fire on radiative forcing depends on the averaging period (Fig. 3B). Averaged over years 0 to 80, net radiative forcing from the different forcing agents was -2.3 ± 2.2 W m^{-2} (Table 1).

A change in fire return times will have consequences for climate forcing (Fig. 3C), based on the time-since-fire trajectories of the different forcing agents estimated from the Donnelly Flats fire, combined with a stand age model (*28*). If the fire return time decreases [as has been suggested from future warming and drying in continental interiors (*29*)], loss of carbon will increase radiative forcing (Fig. 3C). Accounting for all forcing agents, however, leads to a small negative radiative forcing at the global scale (Fig. 3C) and calls into question the positive feedback that has been suggested in past work. The cooling from a decrease in fire return times is likely to be substantially larger in the Northern Hemisphere, taking into account the spatial pattern of the temperature anomalies resulting from the different forcing agents. Specifically, radiative forcing from greenhouse gases has a widely distributed impact on global temperature (*18*), whereas the influence of postfire changes in surface albedo will be concentrated almost entirely in northern regions (*10*, *11*, *13*, *18*).

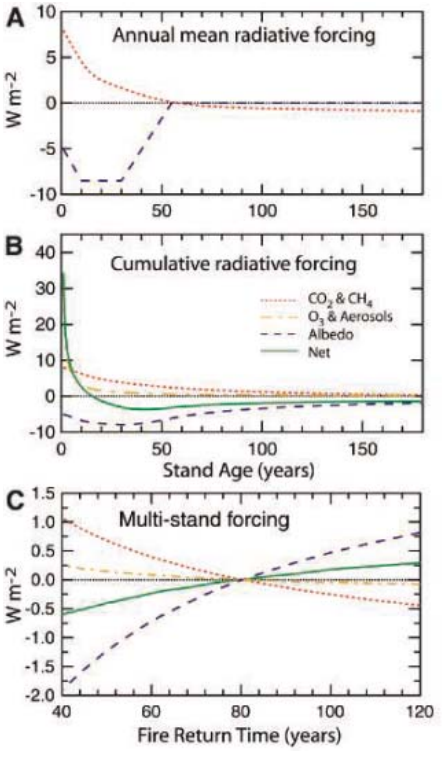

Fig. 3. (**A**) Annual radiative forcing from long-lived greenhouse gases and the postfire trajectory of surface albedo. (**B**) Cumulative annual radiative forcing for the different forcing agents averaged over the time since the fire (or equivalently, the age of the stand). (**C**) Climate forcing of the different components as a function of the fire return time relative to a distribution of stands at steady state with a mean fire return time of 80 years. (C) was constructed with postfire trajectories for the individual agents measured or predicted for the Donnelly Flats fire [e.g., (A)] and the forest stand age distribution model described in the Supporting Online Material. For (C), by definition, each forcing agent had a zero mean at steady state (at a mean fire return time of 80 years).

For the boreal biome as a whole, key factors that are likely to determine the balance between negative and positive radiative forcing associated with fire include burn severity, species establishment in postfire ecosystems, and the duration of winter snow cover. Increased burn severity, for example, may increase aerosol and greenhouse gas emissions, but it is not clear to what extent this may be canceled by greater loss of canopy overstory and consequently higher albedo values during winter and spring. Another unresolved question involves the extent to which fire in Siberian larch forests, which are needleleaf deciduous, has the same influence on postfire surface albedo as reported here for North American needleleaf evergreen forests. Decreases in spring snow cover (*30*) may weaken negative feedbacks associated with postfire increases in surface albedo documented in North America.

Future interactions between the land surface and climate in northern regions may involve both negative feedbacks within the boreal interior (via mechanisms outlined here) and positive feedbacks involving shrub and forest expansion in arctic tundra ecosystems (*31*) and loss of snow cover. Our analysis illustrates how ecosystem processes that generate carbon sources and sinks have inseparable consequences for other forcing agents (*12*, *13*, *32*, *33*). To the extent that the contemporary Northern Hemisphere carbon sink originates from changes in northern forest cover and age (*34*), its value from a climate perspective requires a more nuanced view that encompasses all agents of radiative forcing. Important next steps include reducing uncertainties associated with direct and indirect aerosol effects and disturbance-linked changes in albedo, exploring the combined impacts of feedbacks of the forcing agents estimated here within climate models, and extending this approach to assess the radiative forcing associated with land-cover transitions in temperate and tropical ecosystems.

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Supporting Online Material

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Materials and Methods
SOM Text
Figs. S1 to S10
Tables S1 and S2
References and Notes

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Solar Wind Neon from Genesis: Implications for the Lunar Noble Gas Record

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Lunar soils have been thought to contain two solar noble gas components with distinct isotopic composition. One has been identified as implanted solar wind, the other as higher-energy solar particles. The latter was puzzling because its relative amounts were much too large compared with present-day fluxes, suggesting periodic, very high solar activity in the past. Here we show that the depth-dependent isotopic composition of neon in a metallic glass exposed on NASA's Genesis mission agrees with the expected depth profile for solar wind neon with uniform isotopic composition. Our results strongly indicate that no extra high-energy component is required and that the solar neon isotope composition of lunar samples can be explained as implantation-fractionated solar wind.

Dust grains from both the lunar surface and from meteorites consisting of compacted dust from asteroidal surfaces contain noble gases with isotopic compositions close to those measured in the aluminum foils that trapped solar wind (SW) on the Moon during the Apollo missions (*1*). The SW ions are implanted into these grains during exposure on the lunar or asteroidal surface to depths of up to ~200 nm for SW velocities in the range of 300 to 800 km/s (*2*). Within this implantation zone, deeply sited solar noble gases are substantially enriched in heavy isotopes. Specifically, near the grain surface, $^{20}Ne/^{22}Ne$ ratios are ~13.8 {close to the SW value of 13.7 measured in the Apollo foils [(*1*) and references therein]}, whereas at greater depths, $^{20}Ne/^{22}Ne$ values of solar Ne appear to cluster around 11.2 in many samples (*3*, *4*). This data pattern becomes particularly evident when stepwise in vacuo etching of lunar grains is performed, a technique that releases noble gases from progressively deeper layers (Fig. 1). Some fractionation has always been expected along a depth profile, because the normal SW carries all species at roughly equal speeds, causing heavier isotopes to have somewhat higher energies and greater penetration depths (*5*, *6*). However, this effect will not produce a pronounced clustering near a composition of $^{20}Ne/^{22}Ne = 11.2$ but rather a gradual enrichment of the heavier isotopes with depth (Fig. 1). Because of this fact and the fact that the heavy component in lunar dust was assumed to reside considerably deeper than the several-hundred-nanometer penetration depth of the fastest SW ions (*7*), the heavy component was attributed to solar energetic particles (SEPs) from discrete high-energy events known from in situ measurements in space. Therefore, Ne with a $^{20}Ne/^{22}Ne$ composition of 11.2 was labeled SEP-Ne (*3*, *4*, *8*–*10*). Further support for this distinction was provided by in situ analyses done on the Interplanetary Monitoring Platform (IMP-8) and International Sun-Earth Explorer (ISEE-3) spacecraft of solar energetic particles in the mega–electron volt per atomic mass unit range (*11*, *12*), which indicated $^{20}Ne/^{22}Ne$ ratios distinctly lower than the SW value. In the following discussion, SEP-Ne denotes this hypothetical second solar Ne component in lunar samples, in contrast to solar energetic particles actually measured in situ in space.

SEP-Ne in lunar samples has remained enigmatic. Its inferred abundance relative to the implanted SW is about 25 to 50% (*3*), which is several orders of magnitude higher than expected from long-term averages of present-day fluxes of solar energetic particles in space, even postulating that part of the surface-sited SW got lost (*10*). Thus, the high SEP-Ne abundance suggested a periodic increase in solar activity in the past (*10*, *13*). Furthermore, more recent observations of solar energetic particles on the Advanced Composition Explorer (ACE) spacecraft indicate a highly variable Ne isotopic composition, making it doubtful whether the long-term average solar energetic particle composition differs significantly from the SW value (*14*). Other explanations for the apparent large abundance of SEP-Ne in lunar grains have included a strong depletion of the SW component due to grain-surface sputtering (*10*) and a galactic (*15*) rather than a solar source for the SEP-Ne component; none of these explanations was completely satisfactory.

To investigate this problem, a special bulk metallic glass (BMG) target (*16*, *17*) was exposed to solar particles for 27 months on NASA's Genesis Discovery Mission (*18*). The BMG target lends itself to high-resolution depth profiling of the amount and isotopic composition of implanted solar noble gases by in vacuo online etching (*16*), because it can be etched very homogeneously in nitric acid (*19*). It is one of the few Genesis targets recovered essentially intact.

The depth distribution of SW Ne isotopes was analyzed in two BMG samples of 0.11 and 0.37 cm^2 in 9 and 30 steps, respectively. The Ne isotopic composition became progressively heavier with increasing depth (Fig. 2, fig. S1, and table S1), following a mass-dependent fractionation line. The first minor steps showed $^{20}Ne/^{22}Ne$ ratios around 16, which is distinctly higher than the bulk SW average. With progressive etching, $^{20}Ne/^{22}Ne$ ratios decreased, with a large fraction of the gas displaying ratios around the bulk SW value (Fig. 2). The last steps releasing sufficient gas for precise analyses displayed $^{20}Ne/^{22}Ne$ ratios similar to the SEP-Ne value observed in lunar samples; however, the final point was below the SEP-Ne value. The total Ne released stepwise from the BMG had an average bulk $^{20}Ne/^{22}Ne$ ratio of 13.85 ± 0.11, which is similar within uncertainties to the bulk SW average of 13.75 ± 0.05 as measured by total extraction in three additional BMG samples

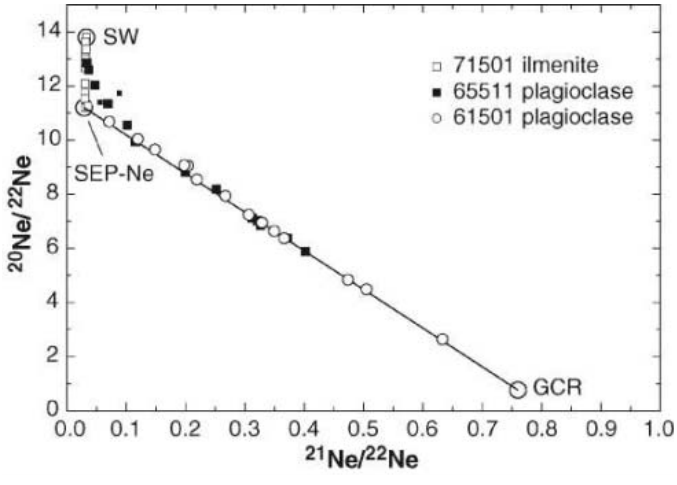

Fig. 1. Ne data from three lunar samples with solar noble gases reported by (*3*, *4*). Gases were released by in vacuo etching, except for sample 61501 [total extraction of samples etched off-line, as explained by (*7*)]. In all samples, besides SW Ne (labeled as SW from the Apollo missions), a second solar component appeared to be present, labeled SEP-Ne. GCR indicates the composition of Ne produced by GCRs as extrapolated from the plagioclase samples.

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(tables S1 and S2). Both values are slightly corrected for backscatter losses (*16*).

Although three of the four largest high-energy solar flare events producing solar energetic particles in solar activity cycle 23 fell into the exposure period of Genesis (*20*), the fluence of these particles was at least three orders of magnitude too small (*13*) to possibly account for the Ne with a SEP-Ne–like isotopic composition (abundance ratio of solar energetic particles/SW < 0.001). Furthermore, in contrast to the much longer–exposed lunar samples, the abundance ratio of deeply and near-surface implanted solar Ne in the metallic glass was unaffected by sputter-induced losses of surface layers. Therefore, the data pattern of the metallic glass in Fig. 2 does not require a mixture of two energetically and isotopically different solar components but rather is due to isotopic fractionation effects upon implantation.

This conclusion is corroborated by ion-implantation modeling using the SRIM (stopping and range of ions in matter) code (*2*, *16*). Apart from the elevated $^{20}Ne/^{22}Ne$ ratios measured in the first few minor steps, the modeled Ne isotopic profile matches the measured profile very well (Fig. 2). In particular, the SEP-Ne value of 11.2 is reached in the modeled profile at around 94 to 97% gas release, which is similar within uncertainties to the measured profile. Progressively lower $^{20}Ne/^{22}Ne$ ratios well below 11.2 are expected in the remaining 3%, although this would be difficult to measure. Therefore, a single SW-Ne component with a uniform isotopic composition over the entire velocity range suffices to explain the depth profile in the BMG.

The data pattern of the BMG is very similar to those of many lunar samples, with two major simplifications. First, Genesis samples do not contain detectable amounts of Ne produced by spallation by galactic cosmic ray particles (GCR-Ne), because no appreciable concentrations of GCR-Ne accumulated during the approximately 3 years in space. This fact is important because a seemingly strong argument for the reality of SEP-Ne had been that in many in vacuo etch runs (such as lunar plagioclase grain separates), the data points of the later etch fractions fall onto a straight line in a Ne three-isotope diagram (Fig. 1). This line passes through the extrapolated GCR point on the lower right and the SEP-Ne point on the upper left, apparently indicating a constant isotopic composition of the SEP-Ne end member over a large depth range. Second, the major gas fractions released early in the BMG etch runs showed higher $^{20}Ne/^{22}Ne$ ratios around 14.3, with the first fraction being as high as 16. In contrast, in the first etching steps, many lunar samples reveal $^{20}Ne/^{22}Ne$ ratios essentially identical to the SW value of ~13.7 determined by the Apollo SW composition experiments. As discussed quantitatively below, the SW-like isotopic composition of gases released from very close to the surface of lunar grains can be explained by the sputtering of grain surfaces due to SW irradiation or mechanical erosion (*5*, *10*, *21*).

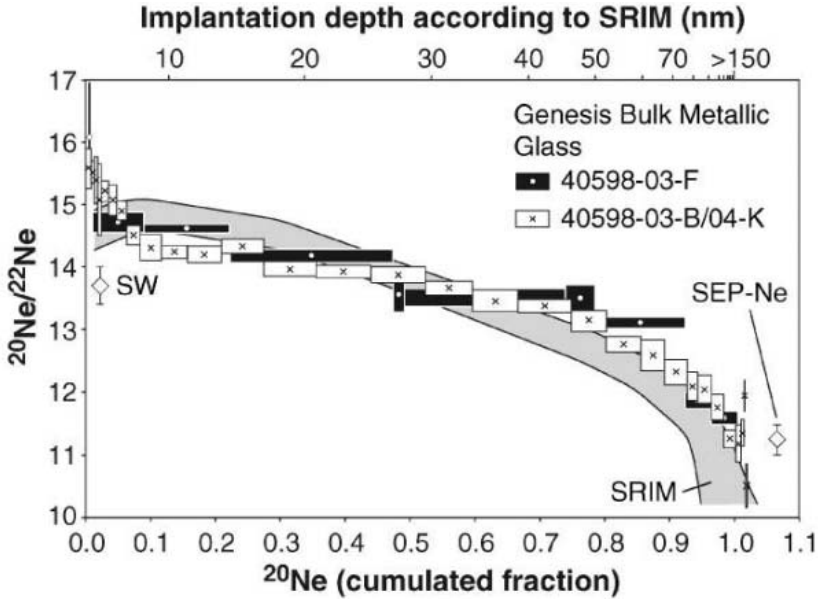

Fig. 2. $^{20}Ne/^{22}Ne$ ratios versus the cumulative fraction of ^{20}Ne released from two in vacuo etch runs of BMG samples (table S1) and SRIM simulations. Total ^{20}Ne concentrations [corrected for backscatter losses of 11.3% (*16*)] were $(1.31 \pm 0.03) \times 10^{12}$ atoms/cm^2 for both runs, similar to the average of $(1.28 \pm 0.09) \times 10^{12}$ atoms/cm^2 deduced from total extraction of three BMG samples used for normalization of the ^{20}Ne fraction (table S2). The length of the bars in the horizontal direction indicates gas amounts released per step; the vertical extension of the bars indicates 2σ uncertainties of $^{20}Ne/^{22}Ne$ ratios, including ion statistics, extraction blank variability, interferences, and mass discrimination. Results from SRIM simulations are plotted as the 2σ uncertainty envelope (gray-shaded). This simulation assumes an implantation of SW with a velocity-independent $^{20}Ne/^{22}Ne$ ratio of 13.75 ± 0.05 (table S2), which is supported by [(*28*) and references therein], with a SW velocity distribution as measured by ACE instruments (*29*). The implantation depth of the measured gas can be approximated from the simulated depth given on the upper *x* axis. Apart from the fact that the high measured values in the first minor steps are not reproduced in SRIM results, simulated and measured $^{20}Ne/^{22}Ne$ profiles agree excellently with each other. Thus, the data pattern of the BMG can be explained by a fractionation of an isotopically uniform SW upon implantation. The SEP-Ne data point is for reference.

To account for the differences between Genesis and lunar exposure conditions, we extended the SRIM simulations (Fig. 3) by implementing surface sputtering, leading to sputter-saturation equilibrium, and GCR-Ne production in a material with plagioclase composition (*22*), with amounts adjusted to match the solar-Ne/GCR-Ne abundance ratio in lunar plagioclase grains (*4*, *16*). At the grain surface, the simulated data points show the true composition of the impinging SW. This corroborates the earlier interpretation that the outermost layers of lunar samples may in fact conserve the true isotopic composition of the SW (*5*, *10*). At greater depths, the simulated mixing curve essentially reproduces the linear trend displayed by many lunar samples. This similarity between measured lunar and simulated data is a very strong indication that the SEP-Ne inferred earlier from the lunar samples actually does not exist. Obviously, fractionated SW-Ne close to the SEP-Ne composition is just abundant enough to be detected in the presence of GCR-Ne, whereas more heavily fractionated SW-Ne at even greater depth becomes too rare to be recognized.

Although discussed repeatedly [as in (*4*, *6*, *23*)], it was difficult for previous authors to appreciate the dominant role of fractionation during implantation because of complications in the exposure history of lunar samples. Thus, the existence of an SEP-Ne component was widely accepted. The realization that SEP-Ne is not needed has become possible primarily thanks to the excellent properties of the BMG target when etched in vacuo, allowing high-resolution depth profiling of implanted noble gases, plus the opportunity to see pure SW-Ne, not mixed with GCR-Ne. Furthermore, the exposure occurred in coordination with excellent monitoring of the SW velocity distributions by in situ measurements with spaceborne instruments. Therefore, exposure conditions were very well constrained, in contrast to the lunar samples. The new explanation of the lunar Ne record has the virtue of not requiring strongly increased solar activity and thus higher solar energetic particle flux, or alternatively an enhanced flux of galactic particles (*15*), in the past. Similar implications can be expected for the other four noble gases and oxygen, for which substantial solar energetic particle–related contributions in lunar samples have been suggested (*10*, *24*). It has been proposed by (*25*) that JEP-nitrogen in lunar ilmenites was isotopically light, opposite to Ne. The interpretation of these data needs to be revisited in light of the present work. Measured isotopic compositions of solar gases heavier than pure SW, rather than implying a prominent con-

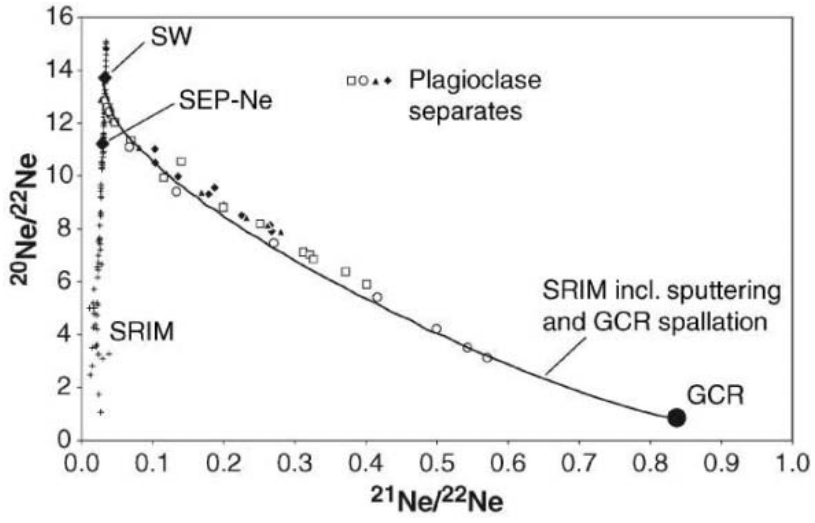

Fig. 3. Comparison of Ne data measured in plagioclase separates from several lunar soils by in vacuo etching (*10*) (squares, circles, triangles, and diamonds), with a simulated mixture of SW Ne fractionated as a function of implantation depth according to SRIM (crosses) and GCR-Ne, assuming SW sputter-saturation equilibrium (*16*). Surface sputtering shifts SRIM results to SW-like values for surface-sited Ne. Adding GCR-Ne moves the modeled data points to higher $^{21}Ne/^{22}Ne$ ratios, leading to a slightly curved line (solid line) that is hardly discernible from the straight line fit through the measured lunar data points for later steps (Fig. 1). Therefore, the lunar data are well reproduced by a SW and GCR mixture only, without an SEP-Ne component. This reflects the fact that the heavily fractionated solar Ne from the deepest layers (with $^{20}Ne/^{22}Ne$ value below that of SEP-Ne) is rare and hardly influences the mixing simulation. This strongly indicates that an independent component with the isotopic composition of SEP-Ne is not required to explain the lunar data.

tribution of solar energetic particles, now indicate a relative enrichment of (isotopically heavy) fractionated SW from greater depths due to, for example, the loss of outermost grain layers. Examples of this are $^{20}Ne/^{22}Ne$ ratios as low as ~11 in interplanetary dust particles (*6*). One may also speculate that the "Ne B" component in Earth's mantle proposed by (*26*, *27*) is possibly a result of a contribution of fractionated implanted SW-Ne during terrestrial accretion.

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Supporting Online Material

www.sciencemag.org/cgi/content/full/314/5802/1133/DC1
Materials and Methods
Fig. S1
Tables S1 and S2
References and Notes

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Sara Endosomes and the Maintenance of Dpp Signaling Levels Across Mitosis

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During development, cells acquire positional information by reading the concentration of morphogens. In the developing fly wing, a gradient of the transforming growth factor–β (TGF-β)–type morphogen decapentaplegic (Dpp) is transduced into a gradient of concentration of the phosphorylated form of the R-Smad transcription factor Mad. The endosomal protein Sara (Smad anchor for receptor activation) recruits R-Smads for phosphorylation by the type I TGF-β receptor. We found that Sara, Dpp, and its type I receptor Thickveins were targeted to a subpopulation of apical endosomes in the developing wing epithelial cells. During mitosis, the Sara endosomes and the receptors therein associated with the spindle machinery to segregate into the two daughter cells. Daughter cells thereby inherited equal amounts of signaling molecules and thus retained the Dpp signaling levels of the mother cell.

In recent years, it has become clear that trafficking through the endosomal pathway is intimately linked with the emission, dispersion, and transduction of intercellular signals during development (*1*). In particular, the endosomal adaptor protein Sara (Smad anchor for receptor activation) plays a key role during transforming growth factor–β (TGF-β) signal transduction (*2*). To study the role of Sara during development, we first determined the

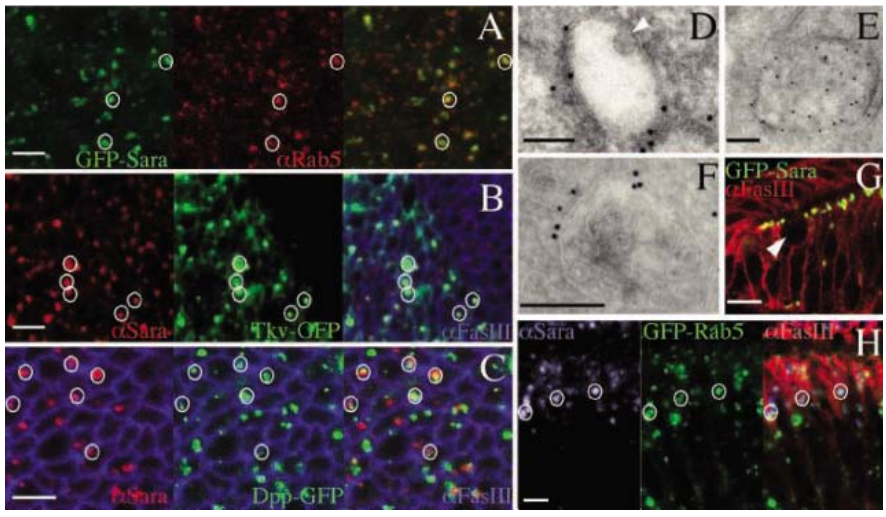

Fig. 1. Sara endosomes in interphase cells. (**A** to **C**) Apical XY confocal sections through imaginal disc cells. (A) Colocalization between Sara endosomes (GFP-Sara, green) and Rab5 (red). [(B) and (C)] Endogenous Sara (red) colocalizes with Tkv-GFP [(B), green] or GFP-Dpp [(C), green]. Cells were outlined by means of FasIII immunostaining (blue). 5.8 ± 0.7% of the total Tkv-GFP is in Sara endosomes and the rest is at the plasma membrane and other vesicular structures (fig. S13C). (**D** to **F**) MVE localization of Tkv-GFP [(D) and (E)] and GFP-Sara (F) with antibodies to GFP in cryoimmunoelectron micrographs. Tkv-GFP is detected at the limiting membrane of an early MVE [(D); arrowhead indicates an invaginating internal vesicle] and at both the limiting membrane and internal vesicles in a mature MVE (E). GFP-Sara is exclusively found at the limiting membrane (F). (**G** and **H**) Z wing cryosections reveal tight apical localization of Sara endosomes [GFP-Sara, (G), green; GFP-Rab5, (H), green; endogenous Sara, (H), blue; FasIII, red]. There is an absence of Sara from more basal Rab5 endosomes (H). Arrowhead in (G) indicates a dividing cell. Examples for colocalization of GFP-tagged Dpp, Tkv, and Rab5 with Sara endosomes are highlighted by white circles in (A) to (C) and (H). Scale bars, 2 μm in (A) to (C) and (H), 100 nm in (D) to (F), and 5 μm in (G).

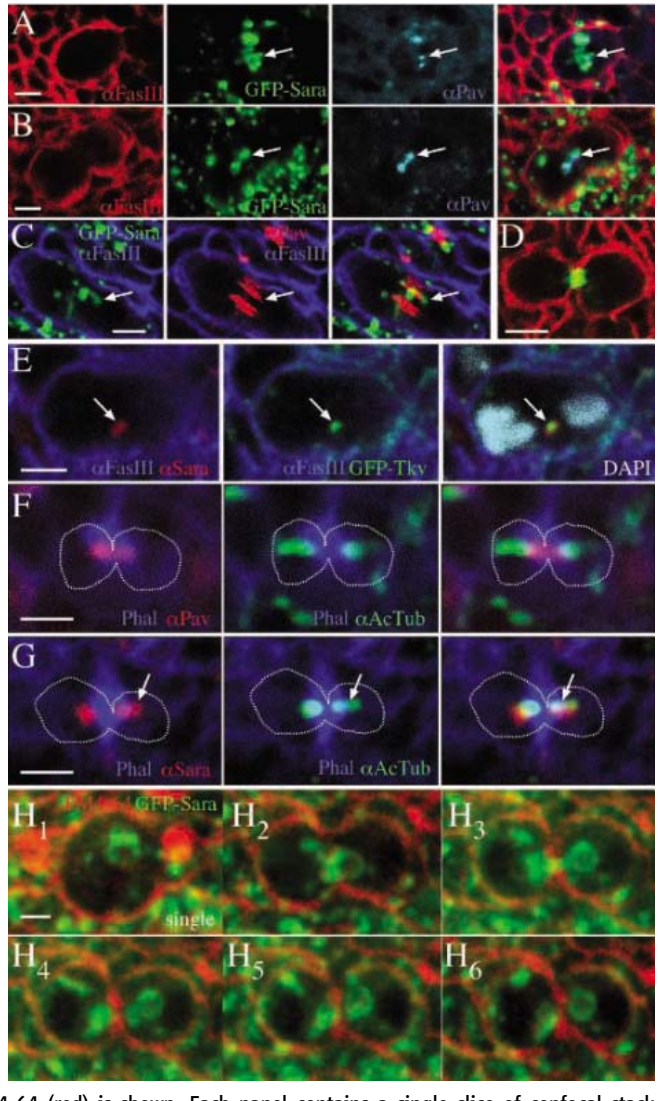

Fig. 2. Sara endosomes in dividing cells. (**A** to **D**) GFP-Sara (green) localizes to the central spindle (arrows) marked by Pavarotti [(A) and (B), blue; (C), red] at progressive stages of mitosis [(A), early anaphase; (C), late anaphase; (B), telophase; (D), cytokinesis] in dividing wing epithelial cells outlined with FasIII [(A), (B), and (D), red; (C), blue]. (**E**) A fraction of intracellular Tkv-GFP (green) colocalizes with endogenous Sara (red) at the cleavage plane (arrows). Cell was outlined by means of FasIII (blue) and DNA was marked by 4′,6′-diamidino-2-phenylindole (DAPI) (light blue). (**F** and **G**) Dividing wing cells stained for Pavarotti [(F), red] or Sara [(G), red], acetylated tubulin (green), and actin with the use of fluorescent phalloidin (blue). Acetylated tubulin appears in the midzone during telophase and surrounds the central spindle defined by Pavarotti (F). At this stage, the Sara endosomes colocalize with acetylated tubulin (G) and not with Pavarotti (arrows). Dotted lines indicate cell outlines. (**H**) Time lapse documenting the segregation of GFP-Sara (green) expressed at high levels. A dividing wing epithelial cell outlined by the lipid dye FM4-64 (red) is shown. Each panel contains a single slice of confocal stack centered on the cleavage site. Scale bars, 2 μm.

subcellular localization of Sara in developing wing epithelial cells in *Drosophila* (*3*). As in mammals (*2*, *4*), Sara in *Drosophila* wing disc cells accumulated at early endosomes. Sara colocalized with the early endosomal marker Rab5 (Fig. 1A and fig. S1E) and with fluorescent dextran internalized in a short pulse (fig. S1, A and E). In contrast, Sara endosomes did not colocalize with dextran after a 40-min chase (fig. S1, B and E). Furthermore, Sara was absent from Rab7-positive late endosomes and lysosomes and Rab11-positive recycling endosomes (fig. S1, C to E). Sara endosomes thus represent an early endosomal compartment.

Sara was targeted to phosphatidylinositol 3-phosphate [PI(3)P]–containing early endosomes via its PI(3)P-binding FYVE domain (figs. S1E and S2, A and B) (*2*) and colocalized with the FYVE-domain protein hepatocyte growth factor–regulated tyrosine kinase substrate (Hrs) (figs. S1E and S2D). Hrs itself colocalizes with early endosomes (*5*), where it initiates multivesicular endosome (MVE) biogenesis (*6*). Like Hrs (*7*) and other FYVE-domain proteins, Sara was found on the MVE-limiting membranes (Fig. 1F and fig. S11), although the internal vesicles show peak levels of PI(3)P (*8*). MVEs can be intermediates in the degradation pathway but have also been implicated in the recycling and storage of endocytic cargo (*9*). Indeed, Sara

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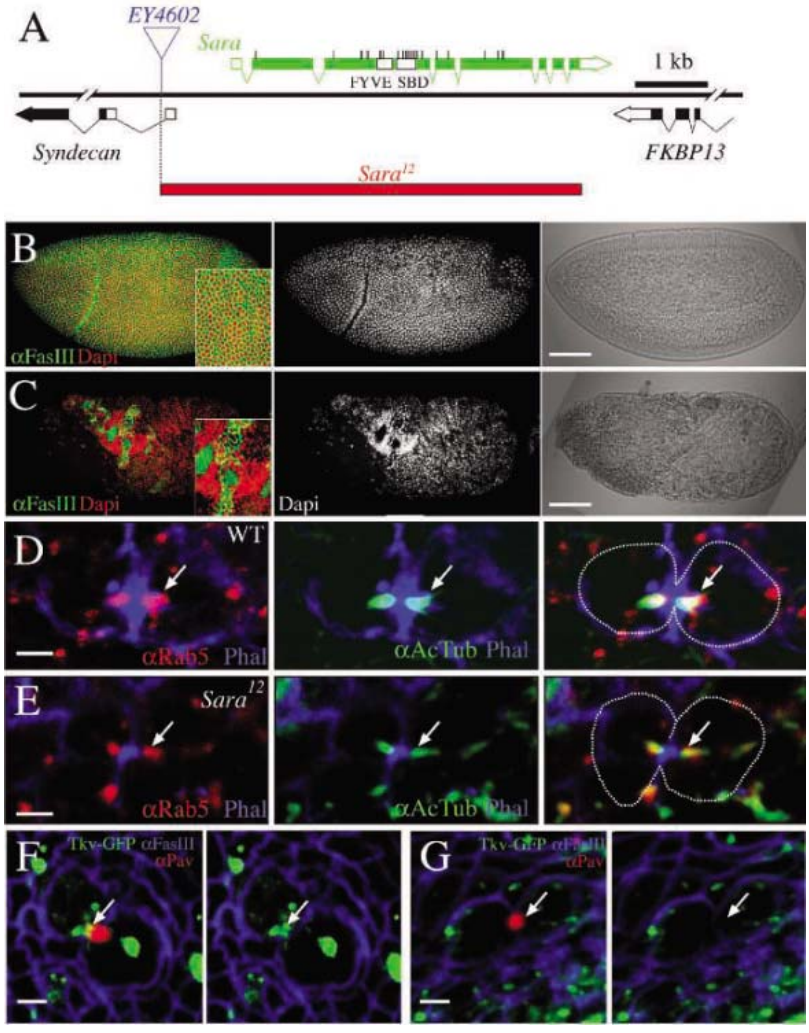

Fig. 3. Sara targets signaling cargo to the central spindle. (**A**) Genomic organization of *Sara* (transcript, green; open reading frame, solid green) and flanking genes (*FKBP13* and *Syndecan*). A transposable element EY4602 (blue) inserted in the first intron of *Syndecan* was used to generate a 5.8-kb deletion by imprecise excision ($sara^{12}$, red), which complements the loss-of-function *Syndecan* alleles. Amino acid exchanges in TILLING mutants, FYVE-, and Smad-binding domains (SBD) are indicated. (**B** and **C**) Double stainings of wild-type (WT) (B) and maternal/zygotic mutant $sara^{12}$ (C) embryos. Cell outlines were marked with FasIII (green) and nuclei were marked by DAPI (red, left panel; gray, center panel). Nomarski images appear in the right panel. High-magnification insets show local cellularization of $sara^{12}$ mutant embryos. The lower density of nuclei in the anterior part of the mutant embryo (C) uncovers a mitotic defect before cellularization. (**D** and **E**) Endogenous Rab5 (red) colocalizes (arrows) with spindle-acetylated tubulin (green) both in WT [(D), 91% of cells] and $sara^{12}$ [(E), 89% of cells] dividing wing disk cells outlined with fluorescent phalloidin (blue). Dotted lines indicate cell outlines. (**F** and **G**) Intracellular Tkv-GFP (green) associates with the midbody marked by Pavarotti (arrows, red) in WT [(F), 90% of cells] but not $sara^{12}$ mutant cells [(G), 10% of cells] outlined by means of FasIII immunostaining (blue). Levels of total Tkv in all endo- and exocytic subcellular compartments in the two daughter cells are approximately equal when the complete *z* axis is quantified (1.20 ± 0.28 ratio of Tkv levels in the sibling cells, $n = 13$ cell pairs; not significantly different from ratio = 1, $P > 0.531$), and overexpression levels do not differ between WT and mutant backgrounds (fig. S13E). Scale bars, 50 μm in (B) and (C) and 2 μm in (D) to (G).

endosomes correspond to a signaling compartment that segregates from the degradation pathway (*10*).

Sara has been implicated in TGF-β and decapentaplegic (Dpp) signal transduction (*2*, *4*, *11*). Consistently, Sara-positive endosomes contained Dpp and its receptor Thickveins (Tkv) (Fig. 1, B and C). Tkv was found at the limiting membrane of early MVEs and the internal vesicles of mature MVEs (Fig. 1, D and E, and fig. S11).

The Sara-positive endosomes were clustered in the top 3 μm of the 30-μm-tall columnar wing epithelium (Fig. 1G and fig. S3, A and C). More basal Rab5-positive early endosomes lacked Sara (Fig. 1H and fig. S3, B and C). In total, only 12% of the Rab5 endosomes were Sara-positive. Thus, Sara defined a strictly apical subpopulation of early endosomes confined to a small volume representing the top 10% of each cell (fig. S3A). We wondered how the mother cells ensure that this polarized compartment and its cargo are partitioned to each daughter cell during mitosis.

Dividing wing cells rounded up at the apical surface of the epithelium during prophase (Fig. 1G). At anaphase, Sara was targeted to the mitotic spindle midzone (also called the central spindle) as monitored by immunostaining with the centraspindlin kinesin Pavarotti/kinesin-like protein–1 (*12*) (Fig. 2, A to C). These Sara-positive structures corresponded to endosomal compartments as they colocalized with internalized dextran and the endosomal markers Rab5 and green fluorescent protein (GFP) fused to a tandem FYVE domain (2xFYVE-GFP) (fig. S4). Furthermore, Sara-positive endosomes at the cleavage plane contained the transmembrane receptor Tkv (Fig. 2E). Later during cytokinesis, the Sara endosomes were displaced off the Pavarotti-positive central spindle and colocalized with acetylated tubulin at both ends of the central spindle (Fig. 2, F and G). In this way, the pool of Sara endosomes targeted to the center of the cell was partitioned into two subpopulations, which then segregated into the two daughter cells. After mitosis, the Sara compartments remained associated with the midbody at the apical side of the epithelial cells (Fig. 2, D and H), which then regained their columnar aspect.

Other endosomal structures, such as lysosomes and late endosomes, did not associate with the spindle but were randomly segregated during mitosis (fig. S5A). Rab11-positive recycling endosomes associate with the centrosomes during cytokinesis (*13*, *14*) (fig. S5B). In the case of Rab5-positive early endosomes, one subset segregated randomly, whereas another population potentially corresponding to the Sara endosomes associated with the central spindle (Fig. 3D and figs. S4B and S5C).

In order to analyze the role of Sara in endosomal or cargo targeting to the central spindle, we generated *Sara* deletions. $sara^{12}$ removes the entire Sara coding region and represents a null mutation (Fig. 3A and fig. S12D). Zygotic $sara^{12}$ mutants died during late larval and pupal development. In rare cases (5%), they survived until adulthood, showing variable vein defects (Fig. 4, L to Q). Most maternal/zygotic $sara^{12}$ mutants (92.2%) died during embryogenesis with erratic phenotypes, including cellularization and dorsal/ventral patterning defects among others (Fig. 3, B and C). Few animals (1.4%; $n = 1520$) reached the third instar larval stage. Both Mad phosphorylation and the pattern of the Dpp target gene *spalt* were normal in the absence of Sara (figs. S10 and S12). Thus, although Sara is essential for activin/TGF-β signaling by recruiting Smad2/3 to the receptor complex for

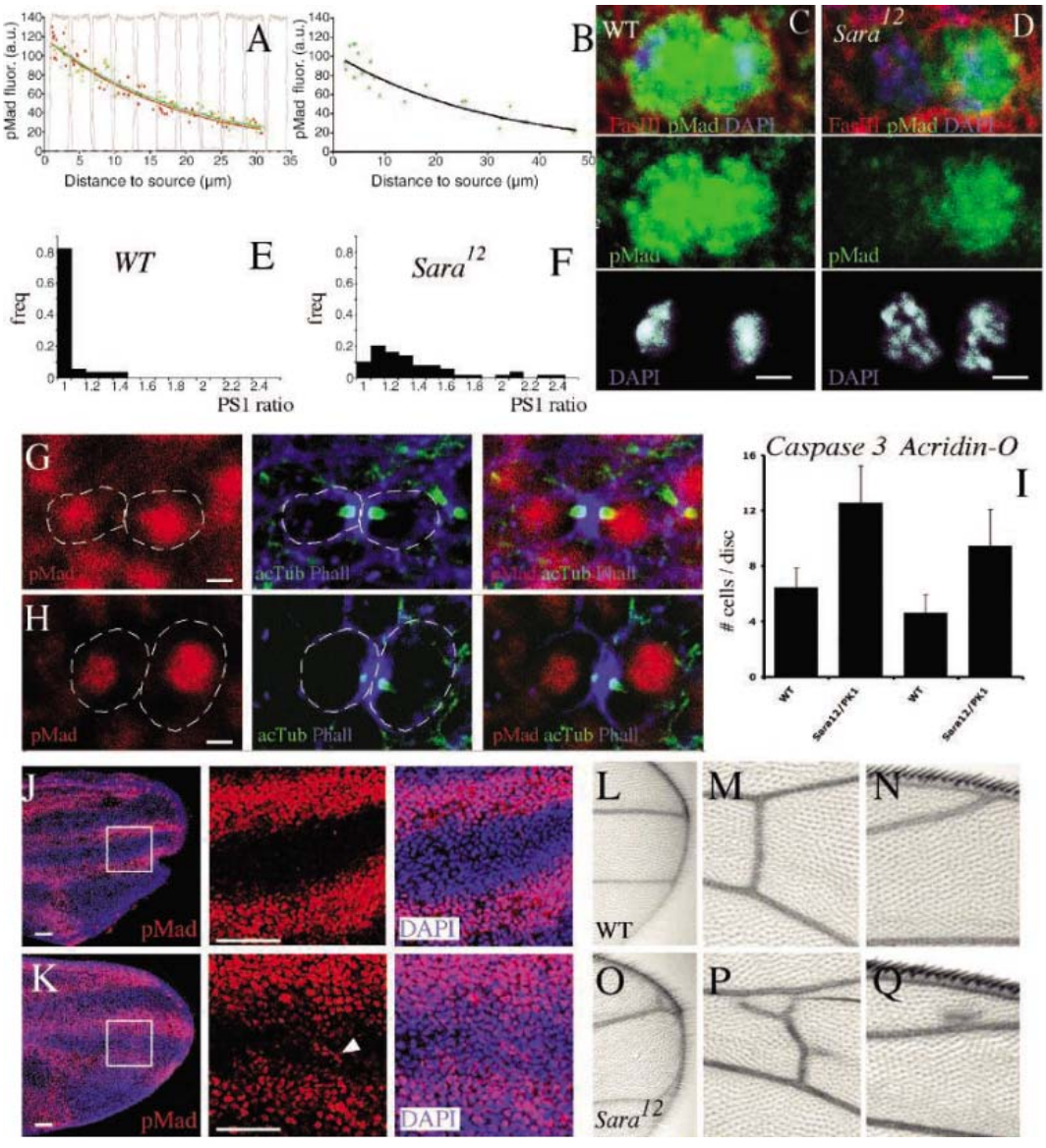

Fig. 4. Sara maintains Dpp signaling levels across mitosis. (**A**) Normalized pMad immunofluorescence intensity in individual cells plotted against the distance from Dpp source. a.u., arbitrary units. Cell outlines correspond to average actual dimensions. The different colors and symbols denote individual disks. (**B**) Normalized pMad signal in mitotic cells versus distance from Dpp source. Although absolute pMad levels are about 10-fold increased relative to interphase, a regular gradient is retained (n = 3 discs). (**C** and **D**) pMad staining (green) in WT (C) and *sara*^{12} mutant sibling cell pairs (D) at the exit of mitosis, counterstained with FasIII (red) and DAPI (blue). Compare equal pMad levels in the WT sibling cells (C) with unequal pMad levels in the mutant (D). (**E** and **F**) Ratios of pMad signals in WT (E) and *sara*^{12} (F) pairs of sibling cells (n = 60 pairs per genotype) at the exit of mitosis. (**G** and **H**) Dividing cells of WT (G) and *sara*^{12}/Df(2R)PK1 (H) pupal wings in 17 hours after puparium formation (apf). Sibling pairs are identified by the accumulation of actin (fluorescent phalloidin, blue) and acetylated tubulin (green) at the contact site. Dashed lines indicate cell outlines. Ratios of pMad (red) intensity between sibling cells are 1.09 for the WT (G) and 2.07 for the mutant pair (H). (**I**) Quantification of apoptosis levels in WT and *sara*^{12}/Df(2R)PK1 wing disks via caspase-3 immunostaining and acridin orange staining. Differences were significant (P < 0.001). Error bars indicate SD. (**J** and **K**) WT (J) and *sara*^{12}/Df(2R)PK1 (K) wings 25 hours apf, stained for pMad (red). Nuclei were marked with DAPI (blue). pMad activation defines the presumptive vein territories, but ectopic pMad (arrowhead) appears in the intervein territory of the mutant wing (center and right panels are magnifications of the boxed areas in the left panels). (**L** to **Q**) *sara*^{12} mutant flies [(O) to (Q)], but not WT control flies [(L) to (N)], exhibit delta formation at vein junctions with the margin [(L) and (O), 85% of mutant wings], branching of the posterior cross-vein [(M) and (P), 28%], and partial vein duplications [(N) and (Q), 15%]. Scale bars, 2 μm in (C), (D), (G), and (H) and 20 μm in (J) and (K).

phosphorylation (*2*, *4*), it is not required in the same way for Dpp/bone morphogenetic protein signal transduction.

We also generated 32 Sara point mutations (Fig. 3A) in two targeted induction of lesions in large genomes (TILLING) reverse genetic screens (*15*). These mutations included *sara*^{1}, a Met^{538}→Lys^{538} amino acid exchange in a conserved position of the FYVE domain that mistargets Sara out of the apical endosomes into the cytosol (fig. S2, E and F). Both *sara*^{12} and *sara*^{1} were lethal mutations sharing phenotypes described below (fig. S6). Both phenotype and lethality could be rescued by expressing a Sara transgene under the tubulin promotor or the GFP-Sara fusion by means of the Gal4 system (table S1 and fig. S7). Thus, the defects described were caused by impaired Sara function, and GFP-Sara was functional.

In both *sara*^{1} and *sara*^{12} zygotic mutants, targeting of endosomes to the central spindle is normal (Fig. 3, D and E, and fig. S8). However, Tkv failed to localize to the central spindle (Fig. 3, F and G, and fig. S13, A, B, and D). Thus, Sara is involved in targeting signaling cargo to endosomes associated with the spindle machinery.

To test whether this biological function of Sara was functionally relevant for Dpp signaling, we quantified Mad phosphorylation in interphase cells and twin daughter cells after cell division. In wild-type wings, phosphorylated Mad (pMad) in interphase cells forms a single exponential concentration gradient profile that parallels the Dpp gradient (*16*): Over a range of 30 μm (~10 cell diameters), the levels of nuclear pMad phosphorylation decayed by a factor of 6 (Fig. 4A and fig. S10). This pMad gradient is thought to encode the anterior/posterior positional information in the disk (*16*).

Mitotic cells showed 10-fold higher levels of Mad phosphorylation than surrounding interphase cells (fig. S9) but retained a pMad gradient profile (Fig. 4B). After mitosis, wild-type twin daughter cells that were identified morphologically or by molecular markers reliably showed equal levels of pMad both in the wing disk (Fig. 4, C and E and fig. S14, B and D) and in pupal wings (Fig. 4G and fig. S15A). In contrast, mutant *sara*^{12} twin daughter cells showed a high variability in pMad levels, reaching up to 2.5-fold

between the twin daughter cells (Fig. 4, D, F, and H, and figs. S14, C and D, and S15B), corresponding to a positional information distance of up to six cells (or 20 μm) (Fig. 4A). Distribution of nonphosphorylated Mad was not affected (fig. S13F). Thus, Sara endosomes store signaling molecules (receptors and/or transcription factors) and are partitioned into the twin daughter cells by association with the central spindle. Sara itself is required for central spindle targeting of the receptor cargo and ensures the maintenance of the levels of pMad within the gradient across mitosis.

The differences in pMad levels between twin sibling cells decrease with time after mitosis in Sara mutants, leading to an overall normal pMad gradient with normal amplitude and range in interphase cells (fig. S10). Consistently, wings of *sara*^{12} escapers did not exhibit large-scale Dpp-related patterning phenotypes (Fig. 4, L to Q). This result implies the existence of mechanisms that smooth the uneven signaling levels in Sara mutants after mitosis. These mechanisms may include the elimination of cells causing discontinuities in the pMad gradient by *brinker*-mediated apoptosis (*17*). Correspondingly, elevated apoptosis levels were observed both in Sara mutant wings (Fig. 4I and fig. S16, B, C, and E) and in Sara mutant clones relative to the surrounding tissue (fig. S16, A and D). Cells entering apoptosis, as monitored by the expression of head involution defective (Hid), exhibited elevated pMad levels in comparison with their neighbors (fig. S16F), which is consistent with previous observations (*18*, *19*). In contrast, daughter cells with inappropriately low signaling levels after mitosis appeared to recover in interphase, implying that, in the larval disk, there is time for de novo signaling by the extracellular gradient of Dpp before cells are committed to a specific fate.

Uneven signaling levels after mitosis should become fixed if differentiation occurred right after division. In the pupal wing, Dpp signaling involved in the determination of vein versus intervein tissue coincides with the final mitotic wave (*20*, *21*). Consistently, we observed cells with ectopically elevated Dpp signaling levels in the intervein territory of Sara mutant wings but not wild-type wings (Fig. 4, J and K), as well as corresponding morphological defects such as branching or partial duplication (Fig. 4, L to Q).

Why do daughter cells retain the Dpp signaling levels of their mother across mitosis, if they are able to read the extracellular gradient of Dpp again after division? We speculate that our observations might be related to the phenomenon of cellular memory of activin signaling in *Xenopus*, where cells remember activation levels for 3 to 6 hours (even across mitosis) (*22*, *23*) because of the retention of activated receptors within the endocytic pathway (*24*). Dpp signaling in flies does not show long-term memory over several cell generations (*25*). However, Dpp signaling may well exhibit short-term memory of a few hours, which would buffer against signaling fluctuations. Uneven distribution of signaling molecules during mitosis (as in Sara mutants) would then generate a situation in which one of the daughter cells may exhibit signaling levels above the levels corresponding to its actual position. Such discontinuities compromise the robustness of the interpretation of the gradient but will to some extent be compensated by *brinker*-dependent apoptosis (*17*). Our observations thus imply a double mechanism to ensure robustness in the interpretation of the gradient: first, error prevention by preempting the appearance of discontinuities after mitosis through Sara and then, should discontinuities appear, error correction through proofreading and apoptosis (*17*, *18*).

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Supporting Online Material

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Materials and Methods
SOM Text
Figs. S1 to S16
Table S1
References

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Abortive Initiation and Productive Initiation by RNA Polymerase Involve DNA Scrunching

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Using single-molecule DNA nanomanipulation, we show that abortive initiation involves DNA "scrunching"—in which RNA polymerase (RNAP) remains stationary and unwinds and pulls downstream DNA into itself—and that scrunching requires RNA synthesis and depends on RNA length. We show further that promoter escape involves scrunching, and that scrunching occurs in most or all instances of promoter escape. Our results support the existence of an obligatory stressed intermediate, with approximately one turn of additional DNA unwinding, in escape and are consistent with the proposal that stress in this intermediate provides the driving force to break RNAP-promoter and RNAP-initiation-factor interactions in escape.

Transcription initiation involves a series of reactions (*1*–*3*). RNA polymerase (RNAP) binds to promoter DNA to yield an RNAP-promoter closed complex (RP_c). RNAP then unwinds ~1 turn of DNA surrounding the transcription start site to yield an RNAP-promoter open complex (RP_o). RNAP then enters into abortive cycles of synthesis and release of short RNA products as an RNAP-promoter initial transcribing complex (RP_{itc}) and, upon synthesis of an RNA product ~9 to 11 nucleotides (nt) in length, escapes the promoter and enters into productive synthesis of RNA as an RNAP-DNA elongation complex (RD_e).

The mechanism by which the RNAP active center translocates in abortive initiation and promoter escape has remained unclear. It has remained unclear because of two seemingly contradictory observations. First, RNA products up to ~8 to 10 nt in length are synthesized in

abortive initiation (*4*–*6*); thus, the RNAP active center translocates relative to DNA in abortive initiation. Second, DNA-footprinting results indicate that the upstream boundary of the DNA segment protected by RNAP is the same in RP_o and in RP_{itc} engaged in abortive synthesis (*7*–*10*); thus, RNAP appears not to translocate relative to DNA in abortive initiation. To reconcile the observation that the RNAP active center translocates in abortive initiation with the observation that RNAP appears not to translocate in abortive initiation, three models have been proposed: (i) "scrunching," which invokes contraction of DNA; (ii) "inchworming," which invokes expansion of RNAP; and (iii) "transient excursions," which invokes transient cycles of forward and reverse RNAP translocations with long intervals between cycles (Fig. 1A) (*4*, *7*, *9*–*12*) [see also proposals for structurally unrelated single-subunit RNAP derivatives (*13*–*19*)].

In previous work, we have developed a single-molecule DNA nanomanipulation approach that detects RNAP-dependent DNA unwinding with ~1–base pair (bp) resolution and ~1-s temporal resolution, and we have applied this approach to detect and characterize RNAP-dependent promoter unwinding upon formation of RP_o (Fig. 1B and fig. S1) (*20*–*22*). In this work, we have applied this approach to test the scrunching model for RNAP-active-center translocation in abortive initiation and promoter escape (Fig. 1A) (*4*, *7*, *11*, *12*). The scrunching model—and only the scrunching model—postulates net changes in RNAP-dependent DNA unwinding during abortive initiation and promoter escape (*23*). Specifically, the scrunching model postulates that RNAP pulls downstream DNA into itself; for each base pair that RNAP pulls into itself, a base pair must be broken and must be maintained broken, and, correspondingly, there must be 1 bp of additional DNA unwinding.

Our first set of experiments addressed abortive initiation occurring in complexes engaged in iterative abortive initiation [complexes prepared with the use of subsets of nucleoside triphosphates (NTPs) insufficient to permit promoter escape and productive initiation]. Our primary experimental system was the T5 N25 promoter, a classic model system for analysis of abortive initiation and promoter escape (*11*) (fig. S2). The initial transcribed sequence of the N25 promoter has no C or G residues in the first 8 nt (fig. S2); therefore, upon preparation of RP_o at the N25 promoter and addition of adenosine triphosphate (ATP) and uridine triphosphate (UTP), one obtains RP_{itc} engaged in iterative abortive synthesis of RNA products up to 8 nt in length ($RP_{itc,\leq 8}$; fig. S2). We also analyzed the N25A5C promoter, a derivative of the N25 promoter that has an altered initial transcribed sequence (fig. S3). With the N25A5C promoter, with appropriate NTP subsets, one obtains RP_{itc} engaged in iterative abortive synthesis of products up to 4 nt in length ($RP_{itc,\leq 4}$) and 8 nt in length ($RP_{itc,\leq 8}$) (fig. S3).

To determine whether scrunching occurs in abortive initiation, we quantified RNAP-dependent DNA unwinding at the N25 promoter in RP_o (0 NTPs) and in $RP_{itc,\leq 8}$ (ATP + UTP) (Fig. 2, A and D). Comparison of the amplitudes of transitions in RP_o and in $RP_{itc,\leq 8}$ in representative single-molecule time traces indicates that the amplitude of transitions is greater in $RP_{itc,\leq 8}$ (Fig. 2A, left). Comparison of the histograms shows clearly that the amplitude of transitions is greater in $RP_{itc,\leq 8}$ (Fig. 2A, right). By combining data obtained with positively supercoiled DNA and data obtained with negatively supercoiled DNA, we can establish unequivocally that there is a difference in DNA unwinding in RP_o and $RP_{itc,\leq 8}$, and we can extract the extent of that difference; namely, upon transition from RP_o to $RP_{itc,\leq 8}$, there is an increase in unwinding of 5 ± 1 bp (Fig. 2D). Consistent with these results, we find that, upon transition from RP_o to $RP_{itc,\leq 8}$ at the N25 promoter, the downstream boundary of the potassium-permanganate-sensitive promoter region shifts from position +2 to position +8, implying an increase in DNA unwinding of 6 bp (fig. S4). We conclude that scrunching occurs in abortive initiation.

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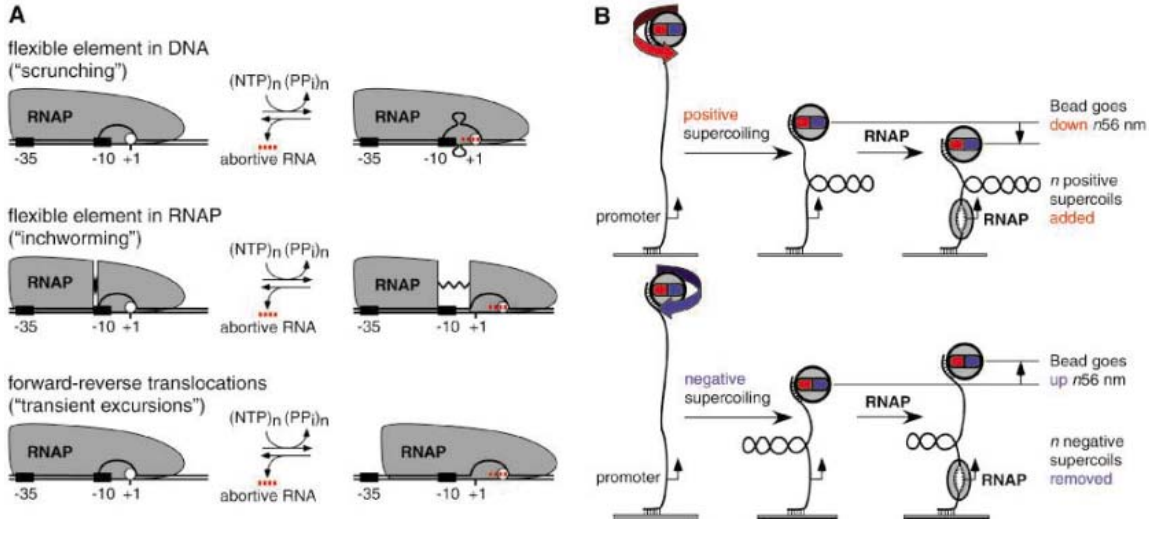


Fig. 1. Background and experimental approach. (**A**) Models for RNAP–active-center translocation in abortive initiation. (Top) The "scrunching" model invokes a flexible element in DNA [(*4*, *7*, *11*, *12*); see also (*13*–*19*)]. In each cycle of abortive initiation, RNAP unwinds downstream DNA and pulls it into itself, accommodating the accumulated DNA as single-stranded bulges in the unwound region; upon release of the abortive RNA, RNAP extrudes the internalized DNA. (Middle) The "inchworming" model invokes a flexible element in RNAP (*9*, *10*). In each cycle of abortive initiation, a module of RNAP containing the active center (white circle) detaches from the remainder of RNAP and translocates downstream; upon release of the abortive RNA, this module of RNAP reverse translocates. (Bottom) The "transient-excursions" model invokes abortive cycles that are transient—too short in lifetime and too infrequent in occurrence to be detected in a time-averaged, population-averaged approach, such as DNA footprinting (*7*). In each cycle of abortive initiation, RNAP translocates downstream as a unit; upon release of the abortive RNA, RNAP reverse translocates as a unit. (**B**) Experimental approach (fig. S1) (*20*–*22*). The end-to-end extension of a mechanically stretched, negatively supercoiled (top) or positively supercoiled (bottom), single DNA molecule containing a single promoter is monitored. Unwinding of *n* turns of DNA by RNAP results in the compensatory loss of *n* negative supercoils or gain of *n* positive supercoils, and a readily detectable, nanometer-scale (*n* times 56 nm), movement of the bead.

To determine whether scrunching requires RNA synthesis, we performed a control experiment in which we provided only the initiating nucleotide, ATP ($RP_{itc,\leq 1}$) (Fig. 2, B and D, and figs. S2 and S5). We also performed a control experiment in which we provided ATP, UTP, and rifampicin, an inhibitor that blocks synthesis of RNA products >2 nt in length ($RP_{itc,\leq 2}$) (Fig. 2, C and D, and figs. S2 and S5). In both experiments, scrunching was not observed (Fig. 2, B and D and C and D). We conclude that scrunching requires RNA synthesis and, more particularly, that scrunching requires synthesis of an RNA product >2 nt in length.

To determine whether the extent of scrunching correlates with RNA length, we quantified RNAP-dependent DNA unwinding at the N25A5C promoter in RP_o (no NTPs), $RP_{itc,\leq 4}$ (ATP + UTP), and $RP_{itc,\leq 8}$ [ATP + UTP+ cytidine triphosphate (CTP)] (Fig. 3, A and B, and figs. S3 and S6). We observed successive, stepwise increases in the amplitudes of transitions (Fig. 3A) and in the corresponding extents of DNA unwinding (Fig. 3B). We observed increases in DNA unwinding of 2 ± 1 bp upon transition from RP_o to $RP_{itc,\leq 4}$ (Fig. 3B) and 5 ± 1 bp upon transition from RP_o to $RP_{itc,\leq 8}$ (Fig. 3B). Within experimental error, the observed increases in unwinding in the preceding experiments and in this experiment agree with the quantitative predictions of the simplest version of the scrunching model, in which increases in unwinding are predicted to equal $N - 2$, where N is the length of the RNA in nucleotides (Fig. 3C). [In the simplest version of the scrunching model, the RNAP active center is predicted to be able to make an RNA product 2 nt in length without translocation, but to need to translocate, and to scrunch, to make longer RNA products; thus, the increase in unwinding is predicted to equal $N - 2$ (Fig. 3C).] We conclude that the extent of scrunching in abortive initiation correlates with RNA length and that it correlates quantitatively as predicted by the simplest model of scrunching.

In each of the preceding experiments, complexes engaged in abortive synthesis and release of RNA products >2 nt in length were observed to be present predominantly in the scrunched state; cycles of transitions between the scrunched state and the unscrunched state having the extent of unwinding in RP_o were not observed (Fig. 2A and figs. S5 and S6). We infer that, at the promoters and saturating NTP concentrations studied, abortive-product synthesis and scrunching are fast relative to abortive-product release and unscrunching, and also are fast relative to the second-scale temporal resolution of our method. Consistent with this inference, recently published results indicate that, at a consensus promoter, at saturating NTP concentrations, the rate-limiting step in abortive initiation is abortive-product release and RNAP-active-center reverse translocation (*24*).

Our results for abortive initiation receive unequivocal support from the companion paper (*25*), which analyzes abortive initiation by means of an independent single-molecule method: single-molecule fluorescence resonance energy transfer (in which scrunching is detected as a decrease in distance between the DNA segments upstream and downstream of the unwound region).

Our second set of experiments addressed promoter escape in complexes engaged in productive initiation (complexes prepared in the presence of all four NTPs). For these experiments, we used DNA constructs having the N25 promoter, followed by a 400- or 100-bp–transcribed region, followed by a terminator (N25-400-tR2 and N25-100-tR2) (fig. S7). These constructs allowed us to monitor complete transcription cycles in the presence of all four NTPs, monitoring promoter unwinding, promoter escape, elongation, and termination, in real time. In addition, as a result of the presence of a terminator, these constructs automatically recycled the DNA molecule, facilitating the collection of large data sets ($n > 100$). (After each transcription cycle, RNAP dissociated from the DNA molecule, rendering the DNA molecule available for the next transcription cycle.) For these experiments, we also used shorter (2 versus 4 kb) DNA molecules (fig. S8) (*22*). This resulted in a decrease in noise and an increase in spatial and temporal resolution.

To determine whether scrunching occurs in productive initiation, we collected and analyzed single-molecule time traces in experiments with

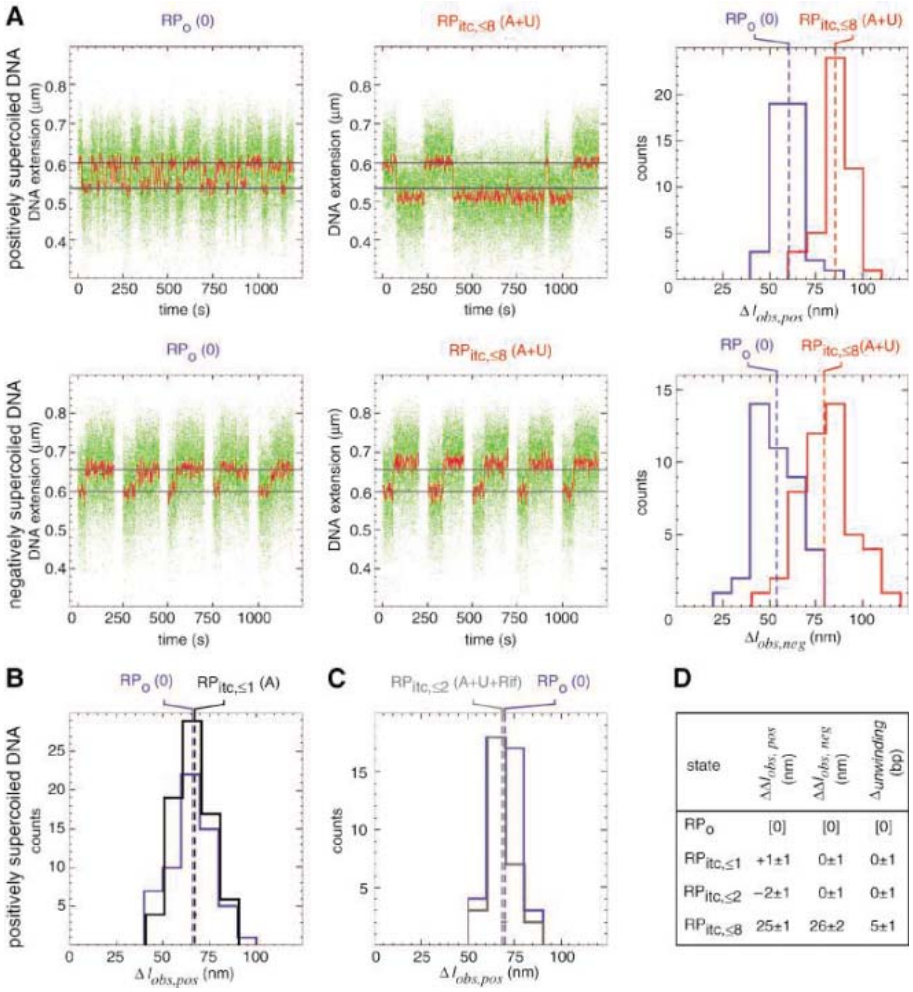


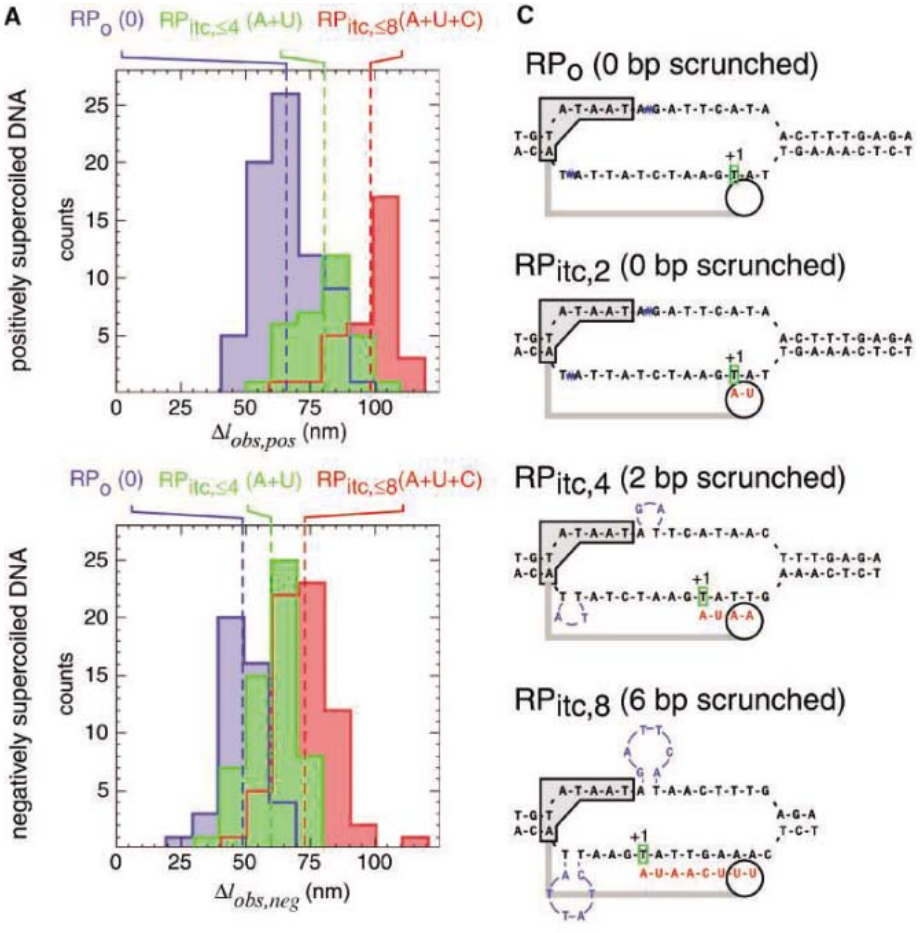
Fig. 2. Scrunching occurs in abortive initiation. (**A**) Single-molecule time traces and transition-amplitude histograms for RP_o (0 NTPs) and $RP_{itc,\leq 8}$ (ATP+UTP) at the N25 promoter. Data for positively and negatively supercoiled DNA are at the top and bottom, respectively. Green points, raw data (30 frames per s); red points, averaged data (1-s window); dashed lines in histograms, means; $\Delta l_{obs,pos}$, transition amplitude with positively supercoiled DNA; $\Delta l_{obs,neg}$, transition amplitude with negatively supercoiled DNA. (**B**) Transition-amplitude histogram for $RP_{itc,\leq 1}$ (from control experiment providing only ATP). (**C**) Transition-amplitude histogram for $RP_{itc,\leq 2}$ (from control experiment providing ATP, UTP, and rifampicin). (**D**) Differences in $\Delta l_{obs,pos}$, $\Delta l_{obs,neg}$, and unwinding relative to values in RP_o (mean ± SE).

N25-400-tR2 and N25-100-tR2 in the presence of all four NTPs (Fig. 4 and fig. S9). Representative single-molecule time traces exhibited series of events, in which each individual event corresponded to a complete transcription cycle, from promoter unwinding through termination (Fig. 4A and fig. S9; events underscored in black). The events were markedly uniform in duration and overall form (Fig. 4A and fig. S9). (Struck by this uniformity, we refer to these time traces as "EKG strips of transcription.") We parsed each event into four unwinding and rewinding transitions: a first transition from the initial state to a state having the extent of unwinding in RP_o; a second transition to a state having the extent of unwinding in the scrunched RP_{itc}; a third transition to a state having an extent of unwinding comparable but not identical to that in RP_o; and a fourth transition returning to the initial state (Fig. 4A and figs. S9 and S10). We assigned these four transitions to the formation of RP_o, the formation of RP_{itc} (with scrunching), the formation of RD_e (with reversal of scrunching), and termination. Scrunching occurred in these events (Fig. 4, A and B, and figs. S9 and S10). Scrunching was manifested as the "overshoot" in unwinding that followed formation of RP_o and preceded formation of RD_e. The observed extent of scrunching was 9 ± 2 bp (Fig. 4B), which agreed, to the base pair, with the predicted extent of scrunching under the $N - 2$ rule (Fig. 3C). [Promoter escape at N25 occurs upon synthesis of an RNA product having a length, N, of 11 nt (fig. S2B).] We conclude that scrunching occurs in promoter escape in productive initiation.

Extensive control experiments documented the assignment of transitions in the preceding paragraph (figs. S11 to S14). When experiments were performed in the absence of NTPs, yielding RP_o, only transition 1 was observed (fig. S11A). When experiments were performed with an NTP subset yielding $RP_{itc,\leq 8}$, only transitions 1 and 2 were observed (fig. S11B). When experiments were performed with an NTP subset yielding a halted elongation complex, $RD_{e,29}$, only transitions 1, 2, and 3 were observed (fig. S11C). When the remaining NTPs were added back to any of the preceding cases, the full process was recapitulated, with full cycles of transcription (fig. S11, A to C). When the terminator was omitted, only transitions 1, 2, and 3 were observed (fig. S12). When the length of the transcribed region was varied, the duration of the phase between transitions 3 and 4 changed, and it changed according to a relationship suggesting an elongation rate of ~10 nt/s [which equals the expected elongation rate for the NTP concentration and temperature (*26–29*)] (figs. S13 and S14). Again, we conclude that scrunching occurs in promoter escape in productive initiation.

To determine whether scrunching occurs in few, many, or all, productive initiation events, we compared the number of transcription cycles that exhibited detectable scrunches to the number of transcription cycles that did not (Fig. 4C). Fully 80% of transcription cycles exhibited a detectable scrunch (Fig. 4C). Thus, most transcription cycles involve scrunching. This percentage, however, represents an underestimate, because the temporal resolution of our approach is insufficient to detect "fast" scrunches (scrunches that have a duration <1 s). From the observed distribution of scrunch lifetimes, we estimate that 20% of scrunches have a duration <1 s (Fig. 4D). Based on the percentage of transcription cycles that exhibit a detectable scrunch (80%) and the estimated percentage of scrunches that are not detected because they have a duration <1 s (20%), it is apparent that ~100% of transcription cycles involve scrunching. We conclude that scrunching occurs in all, or nearly all, transcription cycles. We further conclude that scrunching may be obligatory for promoter escape in productive initiation.

Our overall conclusions are as follows: abortive initiation involves scrunching. Promoter escape involves scrunching. Promoter escape may, and we propose does, involve obligatory scrunching. Promoter escape may, and we propose does, involve an obligatory "stressed



B

state	$\Delta\Delta l_{obs,pos}$ (nm)	$\Delta\Delta l_{obs,neg}$ (nm)	$\Delta_{unwinding}$ (bp)
RP_o	[0]	[0]	[0]
$RP_{itc,\leq 4}$	15±1	10±2	2±1
$RP_{itc,\leq 8}$	33±1	23±2	5±1

Fig. 3. The extent of scrunching correlates with the length of the RNA product. (**A**) Transition-amplitude histograms for RP_o (0 NTPs), $RP_{itc,\leq 4}$ (ATP+UTP), and $RP_{itc,\leq 8}$ (ATP+UTP+CTP) at the N25A5C promoter. Data for positively and negatively supercoiled DNA are at the top and bottom, respectively. (**B**) Differences in $\Delta l_{obs,pos}$, $\Delta l_{obs,neg}$, and unwinding relative to values in RP_o (mean ± SE). (**C**) Prediction of scrunching model: number of base pairs scrunched equals $N - 2$, where N is the length of the RNA product. Sequence-specific RNAP-promoter interactions that define the upstream boundary of the unwound region are indicated by a gray box; RNAP structural elements that constrain the spacing between the upstream boundary of the unwound region and the RNAP active center are indicated by a gray bar; the RNAP active center is indicated by a white circle; the RNA product is in red; position +1 of the template DNA strand is in green; scrunched DNA nucleotides are in blue [(and are positioned as proposed in (*25*)].

intermediate," as originally suggested two decades ago [(*9*); see also (*11*)].

At a typical promoter, promoter escape occurs only after synthesis of an RNA product ~9 to 11 nt in length (*1*–*11*) and thus can be inferred to require scrunching of ~7 to 9 bp ($N-2$, where N = ~9 to 11; Fig. 3C). Assuming an energetic cost of base-pair breakage of ~2 kcal/mol per bp (*30*), it can be inferred that, at a typical promoter, a total of ~14 to 18 kcal/mol of base-pair–breakage energy is accumulated in the stressed intermediate. This free energy is high relative to the free energies for RNAP-promoter interaction [~7 to 9 kcal/mol for sequence-specific component of RNAP-promoter interaction (*1*)] and RNAP-initiation-factor interaction [~13 kcal/mol for transcription initiation factor σ^{70} (*31*)]. We propose that our results demonstrate the existence of an obligatory stressed intermediate, and we propose that the energy accumulated in that obligatory stressed intermediate is the energy that drives the disruption of interactions between RNAP and promoter DNA and between RNAP and initiation factors and, thus, is the energy that drives the transition from initiation to elongation.

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23. The scrunching model postulates that, during abortive initiation, unwinding of downstream DNA base pairs occurs and rewinding of upstream DNA base pairs does not occur, and thus that there is a net increase in DNA unwinding (see length of unwound region in Fig. 1A). In contrast, the inchworming and transient-excursions models postulate that both unwinding of downstream DNA base pairs and rewinding of upstream DNA base pairs occur and thus that there is no net change in DNA unwinding (see length of unwound regions in Fig. 1, B and C).
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Supporting Online Material

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Materials and Methods
Figs. S1 to S15
References

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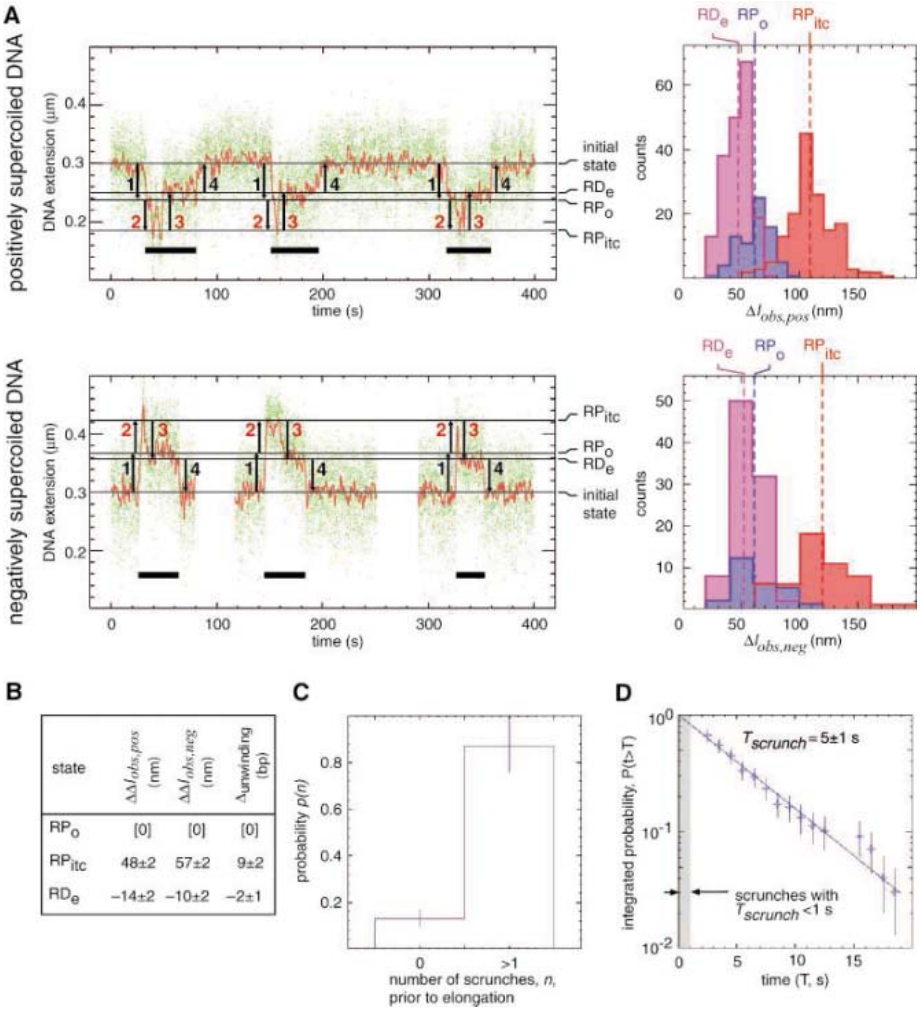

Fig. 4. Scrunching occurs in promoter escape in productive initiation. (**A**) Single-molecule time traces and transition-amplitude histograms for complete transcription cycles on N25-400-tR2 in the presence of all four NTPs. Data for positively and negatively supercoiled DNA are at the top and bottom, respectively. Transcription cycles are indicated by horizontal black bars; unwinding and rewinding transitions are indicated by numbered arrows (red numbered arrows for scrunching and reversal of scrunching); and states are indicated by horizontal lines and labeled on the right. (**B**) Differences in $\Delta l_{obs,pos}$, $\Delta l_{obs,neg}$, and unwinding relative to values in RP_o. (**C**) Fraction of transcription cycles exhibiting at least one detectable scrunch (mean ± SEM; n = 100). (**D**) Distribution of scrunch lifetime [measured from midpoint of transition 2 to midpoint of transition 3 (n = 100)]. Error bars indicate statistical error.

Initial Transcription by RNA Polymerase Proceeds Through a DNA-Scrunching Mechanism

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Using fluorescence resonance energy transfer to monitor distances within single molecules of abortively initiating transcription initiation complexes, we show that initial transcription proceeds through a "scrunching'' mechanism, in which RNA polymerase (RNAP) remains fixed on promoter DNA and pulls downstream DNA into itself and past its active center. We show further that putative alternative mechanisms for RNAP active-center translocation in initial transcription, involving "transient excursions" of RNAP relative to DNA or "inchworming" of RNAP relative to DNA, do not occur. The results support a model in which a stressed intermediate, with DNA-unwinding stress and DNA-compaction stress, is formed during initial transcription, and in which accumulated stress is used to drive breakage of interactions between RNAP and promoter DNA and between RNAP and initiation factors during promoter escape.

Transcription initiation is the first, and the most highly regulated, process in gene expression. In the first steps of transcription initiation, RNAP binds to promoter DNA and unwinds ~14 base pairs (bp) surrounding the transcription start site to yield a catalytically competent RNAP-promoter open complex (RP_o) (*1*–*3*). In subsequent steps of transcription initiation, RNAP enters into initial synthesis of RNA as an RNAP-promoter initial transcribing complex (RP_{itc}), typically engaging in abortive cycles of synthesis and release of short RNA products, and, on synthesis of an RNA product of ~9 to 11 nucleotides (nt), breaks its interactions with promoter DNA, breaks or weakens its interactions with initiation factors, leaves the promoter, and enters into processive synthesis of RNA as an RNAP-DNA elongation complex (RD_e) (*1*–*4*).

The mechanism by which the RNAP active center translocates relative to DNA in initial transcription has remained controversial. DNA-footprinting results indicated that, surprisingly, the upstream boundary of the promoter DNA segment protected by RNAP is unchanged in RP_{itc} as compared with RP_o (*5*–*7*). To reconcile the apparent absence of change in the upstream boundary of the promoter DNA segment protected by RNAP in RP_{itc} with the documented ability of RP_{itc} to synthesize RNA products up to ~9 to 11 nt in length, three models have been proposed (Fig. 1A) (*4*–*8*) [See also proposals for structurally unrelated single-subunit RNAP in (*9*–*15*).]

The first model, termed "transient excursions," invokes transient cycles of forward and reverse translocation of RNAP (Fig. 1A, top) (*5*). According to this model, in each cycle of abortive initiation, RNAP translocates forward as a unit, translocating 1 bp per phosphodiester bond formed [as in elongation; see (*16*)]; on release of the abortive RNA, RNAP reverse-translocates as a unit, regenerating the initial state. According to this model, the cycles of forward and reverse translocation are so short in duration and so infrequent in occurrence that, although they occur, they are not detected by a time-averaged, population-averaged method such as DNA-footprinting.

The second model, termed "inchworming," invokes a flexible element in RNAP (Fig. 1A, middle) (*6*, *7*). According to this model, in each cycle of abortive initiation, a module of RNAP containing the active center detaches from the remainder of RNAP and translocates downstream, translocating 1 bp per phosphodiester bond formed; on release of the abortive RNA, this module of RNAP retracts, regenerating the initial state.

The third model, termed "scrunching," invokes a flexible element in DNA (Fig. 1A, bottom) [(*4*, *5*, *8*); see also (*9*–*15*)]. According to this model, in each cycle of abortive initiation, RNAP pulls downstream DNA into itself, pulling in 1 bp per phosphodiester bond formed and accommodating the accumulated DNA as single-stranded bulges

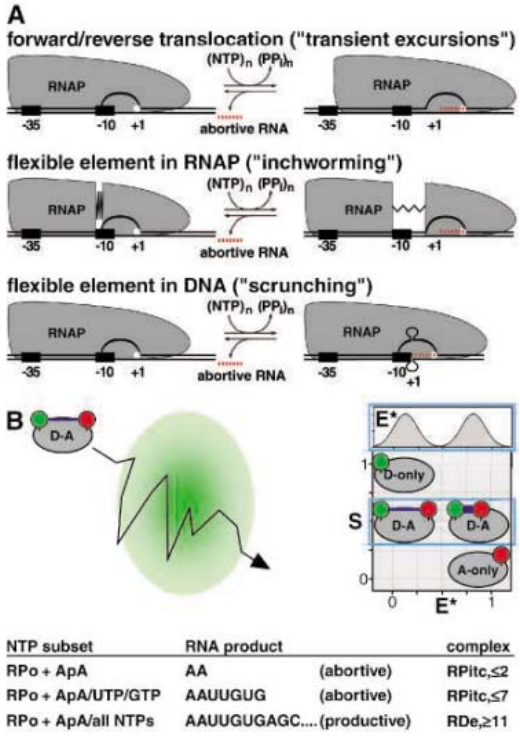

Fig. 1. Background and experimental approach. (**A**) Background. Three models have been proposed for RNAP active-center translocation during initial transcription [(*4*–*8*); see also (*9*–*15*)]: transient excursions, inchworming, and scrunching. White circles, RNAP active center; red dashed lines, RNA; black rectangles: promoter −10 and −35 elements. (**B**) Experimental approach. (Top) Use of confocal microscopy with ALEX (*19*–*21*) to monitor fluorescence of single transcription complexes. Single transcription complexes labeled with a fluorescent donor (D, green) and a fluorescent acceptor (A, red) diffuse through a femtoliter-scale observation volume (green oval; transit time ~1 ms); each molecule is illuminated with light that rapidly alternates between a wavelength that excites the donor and a wavelength that excites the acceptor. For each single molecule, and for each excitation wavelength, fluorescence emission is detected at both donor and acceptor emission wavelengths. This configuration permits calculation of two parameters: the donor-acceptor stoichiometry parameter, *S*, and the observed efficiency of the donor-acceptor energy transfer, *E** (*19*–*21*). The parameter *S* permits identification of molecules containing both donor and acceptor (*S* = 0.4 to 0.9; desired species; boxed in blue), molecules containing only a donor (*S* > 0.9; undesired species, arising from the presence of free σ^{70} molecules and buffer impurities), and molecules containing only an acceptor (*S* < 0.4; undesired species, arising from the dissociation of nonspecific complexes after heparin challenge). Subsequent analysis is performed only on molecules containing both donor and acceptor. (Bottom) Nucleoside triphosphate (NTP) subsets and corresponding RNA products and complexes.

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within the unwound region; on release of the abortive RNA, RNAP extrudes the accumulated DNA, which regenerates the initial state.

The three models are not necessarily mutually exclusive; in principle, combinations of mechanisms may be used, or different mechanisms may be used at different stages of initial synthesis (e.g., one for synthesis of short RNA products, and another for synthesis of longer RNA products).

In this work, we have directly tested the predictions of the three models in Fig. 1A by monitoring distances within single molecules of RP_o and RP_{itc} (*17*). We used fluorescence resonance energy transfer (FRET) (*18*) to monitor distances between fluorescent donors and acceptors incorporated at specific sites within RNAP and DNA. We used confocal optical microscopy with alternating-laser excitation (ALEX) (*19*–*21*) to detect and to quantify fluorescence of single molecules in solution transiting a femtoliter-scale observation volume (Fig. 1B, top left). For each such single molecule, we extracted the donor-acceptor stoichiometry parameter, *S*, and the observed efficiency of donor-acceptor energy transfer, *E** (Fig. 1B, top right). We analyzed *Escherichia coli* RNAP holoenzyme (RNAP core in complex with the initiation factor σ^{70}) (*1*–*3*) and a consensus *E. coli* promoter (*lac*CONS) (*22*) (fig. S1).

We performed four sets of experiments to assess the following: (i) movement of the RNAP leading edge relative to DNA; (ii) movement of the RNAP trailing edge relative to DNA; (iii) expansion and contraction of RNAP; and (iv) expansion and contraction of DNA. In each case, we performed measurements with RP_o containing the initiating dinucleotide ApA [RP_o + ApA, referred to hereafter as RP_o (Fig. 1B, bottom)] and with RP_{itc} engaged in iterative abortive synthesis of RNA products up to 7 nt in length [$RP_{itc,\leq 7}$; prepared by addition of UTP and GTP to RP_o (Fig. 1B, bottom)].

To assess possible movement of the RNAP leading edge relative to downstream DNA in initial transcription, we monitored FRET between a fluorescent donor incorporated at the RNAP leading edge (σ^{70} residue 366, located in σR2, the σ^{70} domain responsible for recognition of the promoter –10 element) and a fluorescent acceptor incorporated at a site in downstream DNA (position +20) (Fig. 2A). The results indicated that, on transition from RP_o to $RP_{itc,\leq 7}$, the mean observed efficiency *E** significantly increases, which implies that the mean donor-acceptor distance, *R*, significantly decreases (Fig. 2A, right). The quantitative increase in mean *E** corresponds to a decrease in mean *R* of ~7 Å (Fig. 2A, right; table S1). Parallel experiments performed using a donor incorporated at a different site at the RNAP leading edge (σ^{70} residue 396, also located in σR2), or using an acceptor incorporated at a different site in downstream DNA (position +15 or position +25), also showed decreases in mean donor-acceptor distance [decreases of ~5 to ~16 Å (fig. S2)]. Control experiments performed in the presence of rifampicin, an inhibitor that blocks synthesis of RNA products >2 nt in length (*23*), showed that the observed decreases in mean donor-acceptor distance required synthesis of RNA products >2 nt in length (fig. S3). We infer that the RNAP leading edge translocates relative to downstream DNA in initial transcription. Furthermore, we infer that, during initial transcription with these reagents and reaction conditions, complexes predominantly occupy states in which the RNAP leading edge is translocated relative to downstream DNA, not states in which the RNAP leading edge is positioned as in RP_o. This implies that the rate-limiting step in initial transcription with these reagents and reaction conditions is abortive-product release and RNAP active-center reverse-translocation [see also (*24*)]. The finding that the RNAP leading edge translocates relative to downstream DNA is consistent with the predictions of all three models for initial transcription (Figs. 1A and 2A). The finding that RNAP predominantly occupies states in which the RNAP leading edge is translocated is incompatible with, or at least problematic for, the transient-excursion model, which invokes translocated states that are short in duration and infrequent in occurrence.

To assess possible movement of the RNAP trailing edge relative to upstream DNA in initial transcription, we monitored FRET between a fluorescent donor incorporated at the RNAP trailing edge (σ^{70} residue 569, located in σR4, the σ^{70} domain responsible for recognition of the

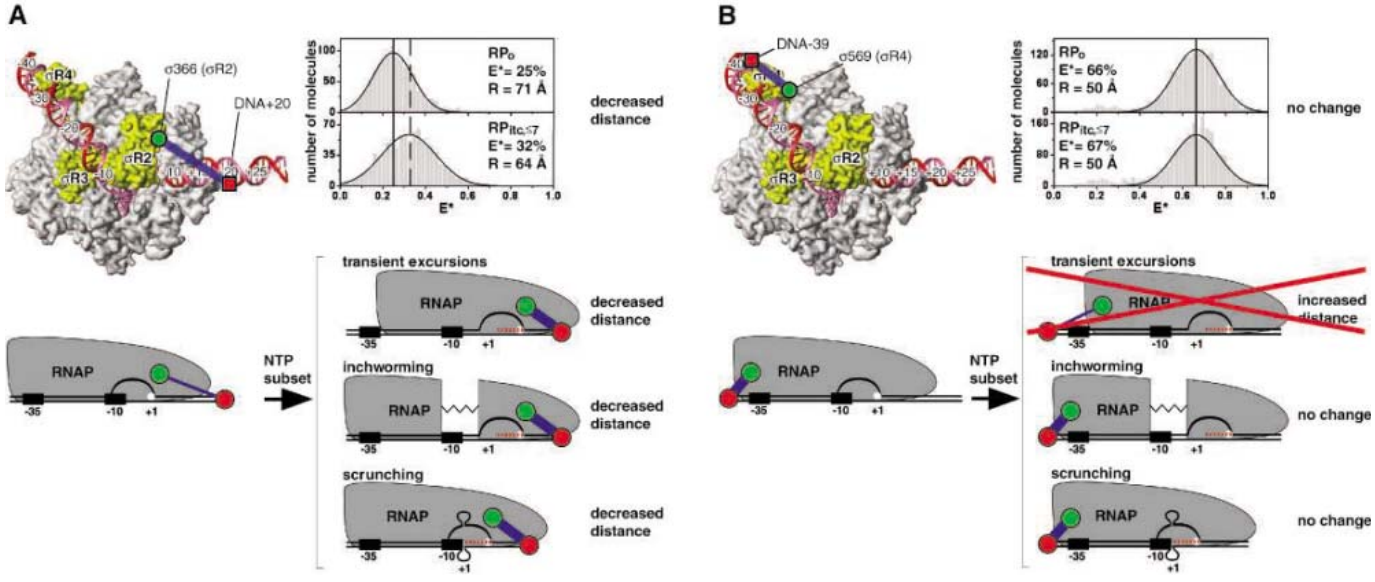

Fig. 2. Initial transcription does not involve transient excursions. (**A**) Experiment documenting movement of the RNAP leading edge relative to downstream DNA [tetramethylrhodamine as donor at σ^{70} residue 366 (located in σR2, the σ^{70} domain responsible for recognition of the promoter –10 element); Cy5 as acceptor at DNA position +20]. (Top left) Structural model of RP_o (*28*) showing positions of donor (green circle) and acceptor (red square). RNAP core is in gray; σ^{70} is in yellow; the DNA template and nontemplate strands are in red and pink, respectively. (Top right) *E** histograms for RP_o and $RP_{itc,\leq 7}$. The vertical line and vertical dashed line mark mean *E** values for RP_o and $RP_{itc,\leq 7}$, respectively. (Bottom) Predictions of the three models. (**B**) Experiment documenting absence of movement of the RNAP trailing edge relative to downstream DNA [tetramethylrhodamine as donor at σ^{70} residue 569 (located in σR4, the σ^{70} domain responsible for recognition of the promoter –35 element); Cy5 as acceptor at DNA position –39]. Subpanels as in (A).

promoter –35 element) and a fluorescent acceptor incorporated at a site in upstream DNA (position –39) (Fig. 2B). In this case, the results indicated that, on transition from RP_o to $RP_{itc,\leq 7}$, mean E^* remains unchanged (Fig. 2B, top right), which implies that the mean donor-acceptor distance remains unchanged. Parallel experiments performed using a donor incorporated at a different site at the RNAP trailing edge (σ^{70} residue 596, also located in σR4) also imply that the mean donor-acceptor distance remains unchanged (fig. S4). Control experiments showed that our probe sites are well positioned to detect a translocation-dependent change in mean donor-acceptor distance and would detect a change if it occurred (fig. S5). We infer that the RNAP trailing edge does not translocate relative to upstream DNA in initial transcription. Specifically, we infer that the σ^{70} domain that interacts with promoter –35 element does not alter its interactions with DNA in initial transcription. This is true even for reaction conditions where it can be shown that the RNAP leading edge translocates relative to downstream DNA (Fig. 2A). These findings are inconsistent with the fundamental prediction of the transient-excursions model; that is, any molecule having the RNAP leading edge translocated relative to DNA also must have the RNAP trailing edge translocated relative to DNA (Figs. 1A and 2B). We conclude that initial transcription does not involve transient excursions.

To assess possible expansion and contraction of RNAP in initial transcription, we first monitored FRET between a fluorescent donor incorporated at the RNAP leading edge (σ^{70} residue 366, located in σR2, and shown in Fig. 2A to translocate relative to downstream DNA) and a fluorescent acceptor incorporated at a site in –10/–35 spacer DNA (position –20)

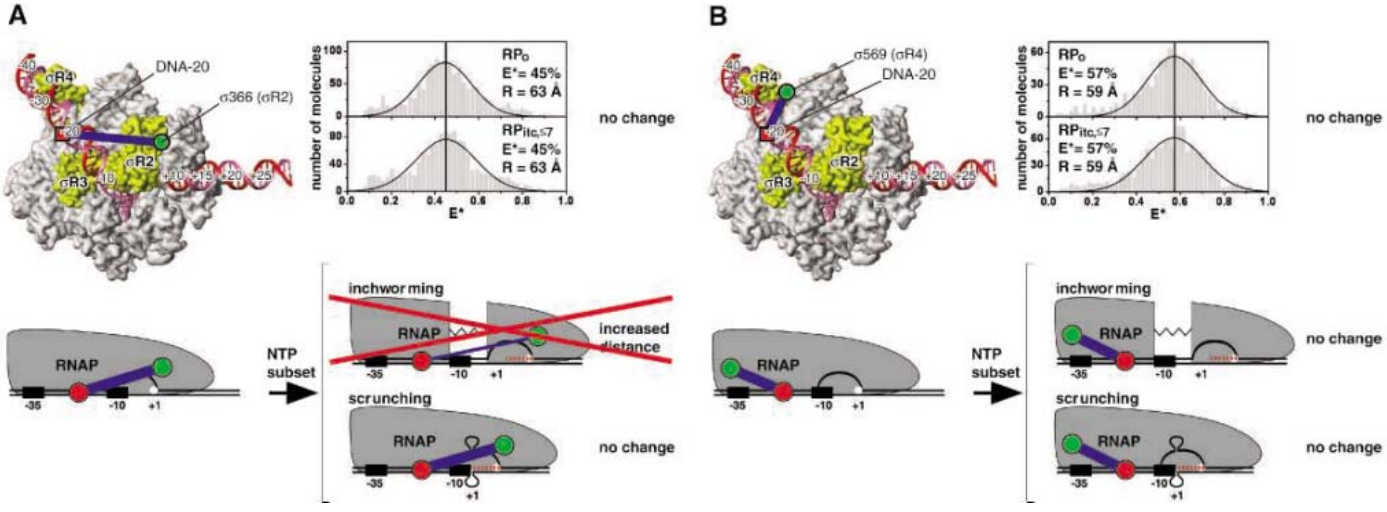

Fig. 3. Initial transcription does not involve inchworming. (**A**) Experiment documenting absence of movement of the RNAP leading edge relative to –10/–35 spacer DNA [tetramethylrhodamine as donor at σ^{70} residue 366 (located in σR2, the σ^{70} domain responsible for recognition of the promoter –10 element); Alexa647 as acceptor at DNA position –20]. Subpanels as in Fig. 2A. (**B**) Experiment documenting absence of movement of the RNAP trailing edge relative to –10/–35 spacer DNA [tetramethylrhodamine as donor at σ^{70} residue 569 (located in σR4, the σ^{70} domain responsible for recognition of the promoter –35 element); Alexa647 as acceptor at DNA position –20]. Subpanels as in Fig. 2A.

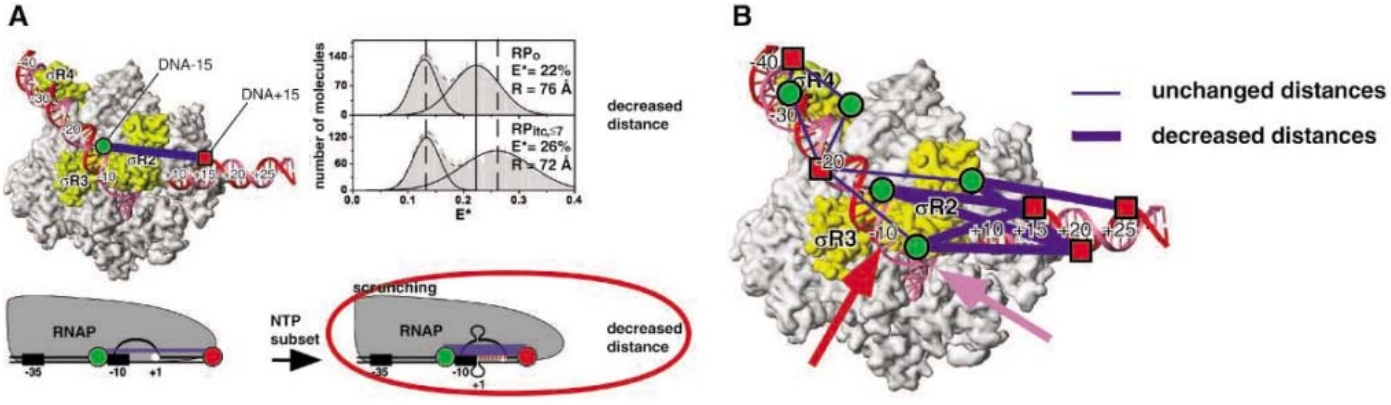

Fig. 4. Initial transcription involves scrunching. (**A**) Experiment documenting contraction of DNA between positions –15 and +15 [Cy3B as donor at DNA position –15; Alexa647 as acceptor at DNA position +15]. Subpanels as in Fig. 2A. [The two donor-acceptor species in the E^* histograms comprise free DNA (lower-E^* species) and RP_o or $RP_{itc,\leq 7}$ (higher-E^* species; higher FRET attributable to RNAP-induced DNA bending)]. Free DNA is present in all experiments, arising from dissociation of nonspecific complexes after heparin challenge during preparation of RP_o, but is detected only in this experiment, because DNA contains both donor and acceptor only in this experiment. (**B**) Summary of results. Structural model of RP_o (*28*) showing all donor-acceptor distances monitored in this work (Figs. 2 to 4A and figs. S2 to S8). Distances that remain unchanged on transition from RP_o to $RP_{itc,\leq 7}$ are indicated with thin blue lines. Distances that decrease on transition from RP_o to $RP_{itc,\leq 7}$ are indicated with thick blue lines. The red and pink arrows show the proposed positions at which scrunched template-strand DNA and scrunched nontemplate-strand DNA, respectively, emerge from RNAP (i.e., near template-strand positions –9 to –10 and near nontemplate-strand positions –5 to –6).

(Fig. 3A). In this case, the results indicated that, on transition from RP_o to $RP_{itc,\leq 7}$, mean E^* remains unchanged (Fig. 3A, top right), which implies that the mean donor-acceptor distance remains unchanged. Parallel experiments performed using a donor incorporated at a different site at the RNAP leading edge [σ^{70} residue 396, also located in σR2, and also shown (in fig. S2) to translocate relative to downstream DNA] also imply that the mean donor-acceptor distance remains unchanged (fig. S6). We next monitored FRET between a fluorescent donor incorporated at the RNAP trailing edge (σ^{70} residue 569 or σ^{70} residue 596, located in σR4) and a fluorescent acceptor incorporated at a site in the –10/–35 spacer DNA (position –20) (Fig. 3B and fig. S6). In this case also, the results indicated that, on transition from RP_o to $RP_{itc,\leq 7}$, mean E^* remains unchanged (Fig. 3B and fig. S6), which implies that the mean donor-acceptor distance remains unchanged. We infer that RNAP does not expand or contract in initial transcription. Furthermore, we infer that the leading edge of RNAP does not translocate relative to DNA upstream of the unwound region—even under reaction conditions where we have shown that the RNAP leading edge translocates relative to downstream DNA (Fig. 2A). These findings are inconsistent with the fundamental prediction of the inchworming model: namely, any molecule having the RNAP leading edge translocated relative to downstream DNA also must have the RNAP leading edge translocated relative to DNA upstream of the unwound region (Figs. 1A and 3A). We conclude that initial transcription does not involve inchworming.

To assess possible expansion and contraction of DNA in initial transcription, we monitored FRET between a fluorescent donor incorporated at a site in –10/–35 spacer DNA (position –15) and a fluorescent acceptor incorporated at a site in downstream DNA (position +15) (Fig. 4A). The results indicated that, upon transition from RP_o to $RP_{itc,\leq 7}$, mean E^* significantly increases (Fig. 4A, top right), which implies that the mean donor-acceptor distance, mean R, significantly decreases. The quantitative increase in mean E^* corresponds to a decrease in mean R of ~4 Å (Fig. 4A, top right). Additional experiments performed using an acceptor incorporated at a different site in downstream DNA (position +20) also showed a decrease in mean donor-acceptor distance (decrease of ~6 Å; fig. S7). Control experiments performed in the presence of rifampicin showed that the observed decreases in mean donor-acceptor distance required synthesis of an RNA product >2 nt in length (fig. S8). We infer that the DNA segment between –10/–35 spacer DNA and downstream DNA contracts in initial transcription. These findings document the fundamental prediction of the simplest version of the scrunching model; that is, any molecule having the RNAP leading edge translocated relative to downstream DNA also must have contraction—scrunching—of the DNA segment between –10/–35 spacer DNA and downstream DNA (Figs. 1A and 4A). We conclude that initial transcription involves scrunching.

We note that all measured distances between RNAP and upstream DNA or –10/–35 spacer DNA remain unchanged on transition from RP_o to $RP_{itc,\leq 7}$, (Fig. 4B, thin blue lines), whereas all measured distances between RNAP and downstream DNA, or between the –10/–35 spacer and downstream DNA, decrease on transition from RP_o to $RP_{itc,\leq 7}$, (Fig. 4B, thick blue lines). We infer that DNA scrunching occurs exclusively within the DNA segment comprising positions –15 to +15. This DNA segment contains the unwound region ("transcription bubble") and, in structural models of RP_o (*22*, *25–28*), is located within and immediately upstream of the RNAP active-center cleft. Inspection of structural models of RP_o and RD_e (*22*, *25–29*) indicates that there is insufficient space within the RNAP active-center cleft to accommodate scrunched DNA and that scrunched DNA instead must emerge from RNAP into bulk solvent immediately upstream of the RNAP active-center cleft. Although the locations at which scrunched DNA emerges are not known, we propose that the scrunched template DNA strand and nontemplate DNA strand emerge at or near the points where the respective DNA strands normally emerge from RNAP immediately upstream of the RNAP active-center cleft; that is, at or near positions –9 to –10 of the template strand and positions –5 to –6 of the nontemplate strand (Fig. 4B, red and pink arrows).

The results in this paper and in the companion paper (*30*) establish that initial transcription involves DNA scrunching. In contrast, processive transcription elongation involves simple translocation, not DNA scrunching [(*16*); see also (*19*)]. Thus, there is a fundamental difference in the mechanisms of RNAP active-center translocation in initial transcription and processive transcription elongation.

The results in this paper and in the companion paper (*30*) provide strong support for existence of a "stressed intermediate" in initial transcription (*4*, *6*), specifically a stressed intermediate with accumulated DNA-scrunching stress (DNA-unwinding stress and DNA-compaction stress). We postulate that the accumulated DNA-scrunching stress in the stressed intermediate provides the driving force for abortive initiation and also provides the driving force for promoter escape and productive initiation. Thus, we postulate that the accumulated DNA-scrunching stress in the stressed intermediate can be resolved in two ways: either (i) by releasing the RNA product, retaining interactions with promoter DNA, retaining interactions with initiation factors, retaining an unchanged position of the RNAP trailing edge, extruding scrunched DNA, and re-forming RP_o (abortive initiation); or (ii) by retaining the RNA product, breaking interactions with promoter DNA, breaking interactions with initiation factors, translocating the RNAP trailing edge, and forming RD_e (promoter escape and productive initiation).

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Materials and Methods
Figs. S1 to S8
Table S1
References

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N-Linked Glycosylation of Folded Proteins by the Bacterial Oligosaccharyltransferase

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N-linked protein glycosylation is found in all domains of life. In eukaryotes, it is the most abundant protein modification of secretory and membrane proteins, and the process is coupled to protein translocation and folding. We found that in bacteria, N-glycosylation can occur independently of the protein translocation machinery. In an in vitro assay, bacterial oligosaccharyltransferase glycosylated a folded endogenous substrate protein with high efficiency and folded bovine ribonuclease A with low efficiency. Unfolding the eukaryotic substrate greatly increased glycosylation. We propose that in the bacterial system, glycosylation sites are located in flexible parts of folded proteins, whereas the eukaryotic cotranslational glycosylation evolved to a mechanism presenting the substrate in a flexible form before folding.

The addition of N-linked oligosaccharides to proteins is a vital process in eukaryotic cells, involved in, for example, control of protein folding, sorting and stability (*1*, *2*), cell-cell and cell-virus interactions (*3*), and host-immune responses (*4*, *5*). Synthesis of glycoproteins occurs in the lumen of the endoplasmic reticulum (ER) where a multisubunit enzyme, oligosaccharyltransferase (OTase), transfers a $Glc_3Man_9GlcNAc_2$ moiety from a lipid-pyrophosphate donor to selected asparagines within nascent polypeptide chains (*6*). *N*-Glycan addition occurs on polypeptides entering the ER through the translocation pore, a process referred to as cotranslocational glycosylation. Because the acceptor protein is translocated in an extended conformation, the OTase is thought to recognize polypeptides in an unfolded state (*7*, *8*). Consistent with this model, the OTase has been found to interact with the Sec translocase in vivo (*9*). However, for some proteins, glycosylation can occur after the entire polypeptide has been translocated into the ER (posttranslocational glycosylation) (*10*), potentially while folding is still incomplete.

The homologous biosynthetic pathway of N-linked glycoproteins in bacteria is less complex. The glycosylation machinery of *Campylobacter jejuni* is encoded by a single gene cluster called *pgl* (protein glycosylation) (*11*), sufficient for recombinant protein glycosylation in *Escherichia coli* (*12*). The *C. jejuni* OTase, PglB, is a single, integral membrane protein with high sequence similarity to the catalytic subunit of the eukaryotic OTase, STT3. Like its eukaryotic counterpart, PglB transfers oligosaccharides ($GalNAc_5GlcBac$, where Bac is 2,4-diacetamido-2,4,6-trideoxy-D-glucopyranoside) from a lipid-pyrophosphate donor to asparagine side chains within proteins (*11*). The consensus sequence recognized on the polypeptide is also similar between the two systems (D-X-N-Z-S/T and N-X-S/T for the bacterial and eukaryotic systems, respectively, where X and Z are any amino acid except proline) (*13*, *14*). The N-X-S/T motif, as well as the requirement for a 2′-acetamido group at the reducing end (*15*), are thought to play integral roles in catalysis in both eukaryotic and prokaryotic OTases.

To determine if bacterial glycosylation was coupled to translocation as in the eukaryotic system, we analyzed the time course of glycosylation for an endogenous model *C. jejuni* glycoprotein, AcrA, by pulse-chase experiments (Fig. 1). Cells expressing the functional *pgl* locus from *C. jejuni* and AcrA containing a Sec-dependent signal peptide (ssPelB) (*12*) were metabolically labeled with a short pulse of

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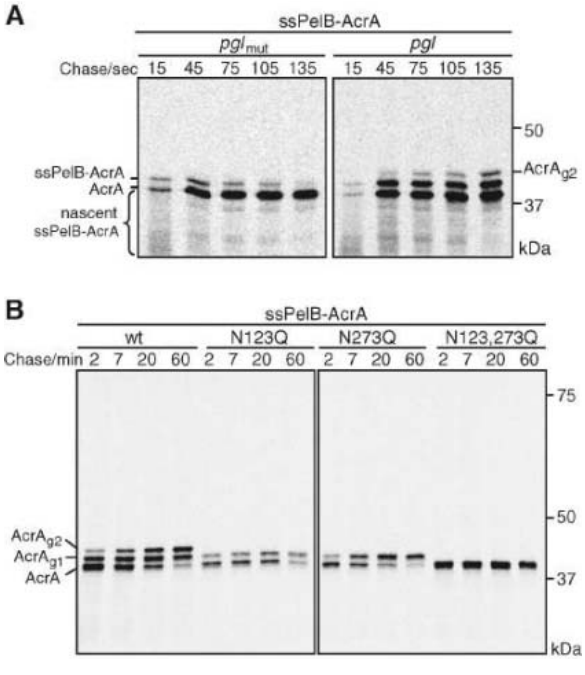

Fig. 1. Kinetics of bacterial N-glycosylation of AcrA in vivo. *E. coli* cells were labeled with ^{35}S-methionine for 15 s (A) or 30 s (B) and chased for the indicated times with nonlabeled methionine. (**A**) Autoradiographs of SDS-PAGE of immunoprecipitated AcrA expressed in glycosylation-competent (*pgl*) and -deficient (pgl_{mut}) cells. The positions of unprocessed (ssPelB), monoglycosylated (g1), and diglycosylated (g2) AcrA are indicated. (**B**) Glycosylation after completion of signal-peptide cleavage from 2 to 60 min. AcrA proteins containing two (wild type), one (N123Q or N273Q), or no glycosylation (N123Q, N273Q) sequons were expressed in glycosylation-competent cells and analyzed as in (A).

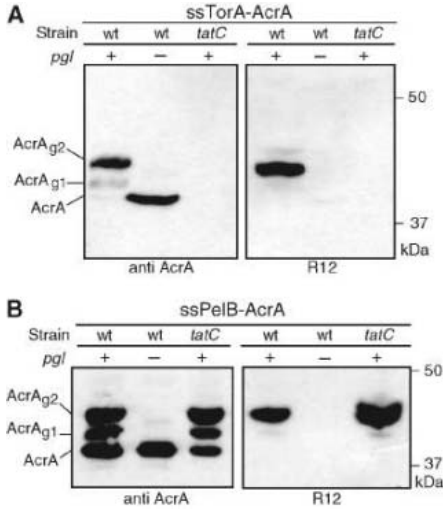

Fig. 2. N-Glycosylation is independent of the translocation process. Periplasmic extracts derived from cells expressing (**A**) ssTorA-AcrA fusion protein or (**B**) ssPelB-AcrA fusion protein, in glycosylation-competent (+), glycosylation-deficient (–), and wild-type (wt) or *tatC* cells, were separated by SDS-PAGE, immunoblotted, and analyzed with antiserum raised against AcrA and with the R12 serum predominantly reactive toward the oligosaccharide modification (*12*).

^{35}S-methionine followed by addition of an excess of nonlabeled methionine (chase). AcrA in glycosylation-deficient cells (Fig. 1A) showed a single band after 135 s, suggesting that AcrA was synthesized and the signal peptide processed. The same was true for AcrA with both glycosylation sequons mutated (N123Q and N273Q) (Fig. 1B). At an equivalent time point, three forms were detected when the wild-type AcrA protein with two glycosylation sites was analyzed (Fig. 1A). The two upper bands were the mono- and diglycosylated AcrA. The intensity of the doubly glycosylated form increased over time, suggesting a continuous increase in glycosylation per molecule over the entire 60 min of the chase (Fig. 1B). Glycosylation continued long after signal sequence cleavage, suggesting posttranslocational glycosylation. This was also observed in AcrA point mutants with only a single glycosylation site.

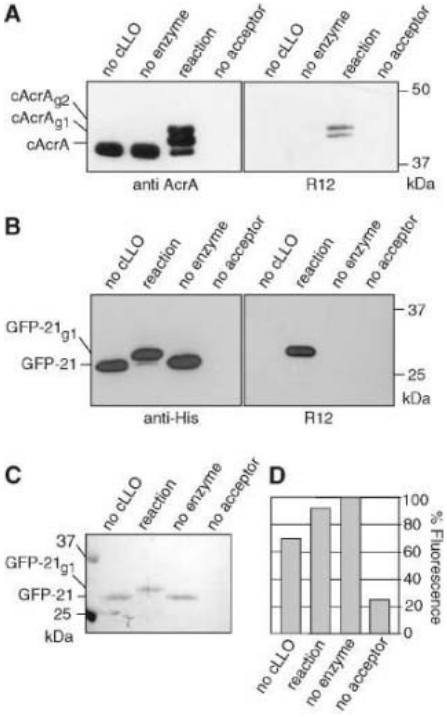

Fig. 3. In vitro glycosylation of recombinant AcrA and GFP-21. Cytoplasmically expressed AcrA (cAcrA) (**A**) or GFP-21 (**B**) were used as acceptor proteins and in vitro glycosylated. Reaction products were separated by SDS-PAGE, immunoblotted, and analyzed with antiserum raised against AcrA and with the R12 serum predominantly reactive toward the oligosaccharide modification (*12*). The position of monoglycosylated (g1) and diglycosylated (g2) protein is indicated. Controls omitting either PglB ("no enzyme"), cLLO, or the acceptor protein are shown. Reactions from (B) were Coomassie stained (**C**), or their fluorescence was measured (**D**) (*19*). Relative fluorescence intensities are indicated. The high signal in the "no acceptor" control is likely an artifact due to light scattering by the cLLO.

The posttranslocational progression of AcrA glycosylation suggested a translocation-independent reaction. To address the hypothesis, we exchanged the PelB signal sequence of the AcrA protein by the TorA signal (ssTorA), directing the fusion protein to the twin-arginine translocation (TAT) machinery that is able to transport folded and oligomeric proteins across membranes (*16*–*18*). The ssTorA-AcrA fusion protein was expressed in *E. coli* cells harboring the *pgl* or pgl_{mut} locus. Periplasmic, glycosylated AcrA was only detected in glycosylation- and TAT-competent cells (Fig. 2A). Mass spectrometric (MS) analysis of the putative doubly glycosylated band (AcrA$_{g2}$) confirmed that the same positions as those in the protein translocated by the Sec-system were N-glycosylated (N123 and N273) (fig. S1). In cells with an inactive TAT-secretion system due to a *tatC* mutation (*16*), the complete absence of AcrA in periplasmic extracts (Fig. 2A) demonstrated that ssTorA-AcrA translocated exclusively via the TAT and not the Sec system. ssPelB-AcrA transported via the Sec system was glycosylated to a similar extent in both the wild-type and *tatC* mutant (Fig. 2B). Thus, N-glycosylation was uncoupled from the translocation machinery in our experimental system, and PglB was able to glycosylate folded proteins exported via the TAT machinery.

To address the folding state of acceptor proteins during glycosylation, we used an in vitro glycosylation assay. PglB was expressed in *E. coli* cells and purified from solubilized membrane fractions (*19*). We then incubated PglB with cytoplasmically expressed and purified AcrA and a crude lipid extract (cLLO) of *E. coli* cells synthesizing the *C. jejuni* LLO by the pgl_{mut} locus (*20*). SDS–polyacrylamide gel electrophoresis (PAGE) and immunoblotting revealed the presence of glycosylated AcrA (Fig. 3A). MS analysis of AcrA with higher molecular weight confirmed the modification of N123 and N273 in this product by the *C. jejuni* oligosaccharide. Thus, PglB was able to glycosylate an independently expressed and purified AcrA in vitro at multiple sites on the same molecule. Eukaryotic OTases, by contrast, do not possess any in vitro activity for folded polypeptides (*21*).

To investigate the folding state of the acceptor protein during glycosylation directly, we grafted a 21–amino acid sequence containing the AcrA N123 glycosylation site into an insertion-tolerant loop present in the green fluorescent protein (GFP) (*22*). This GFP-derivative (GFP-21) was tested for accepting an N-glycan in our in vitro glycosylation assay (*20*). A higher molecular weight protein that was observed to cross-react with the glycosylation-specific serum band, GFP-21, was glycosylated to >90% at the single site in this reaction (Fig. 3C). The shift was due to a glycan attachment to the glycosylation site present in the grafted AcrA-derived loop (fig. S2). Moreover, the fluorescence of the

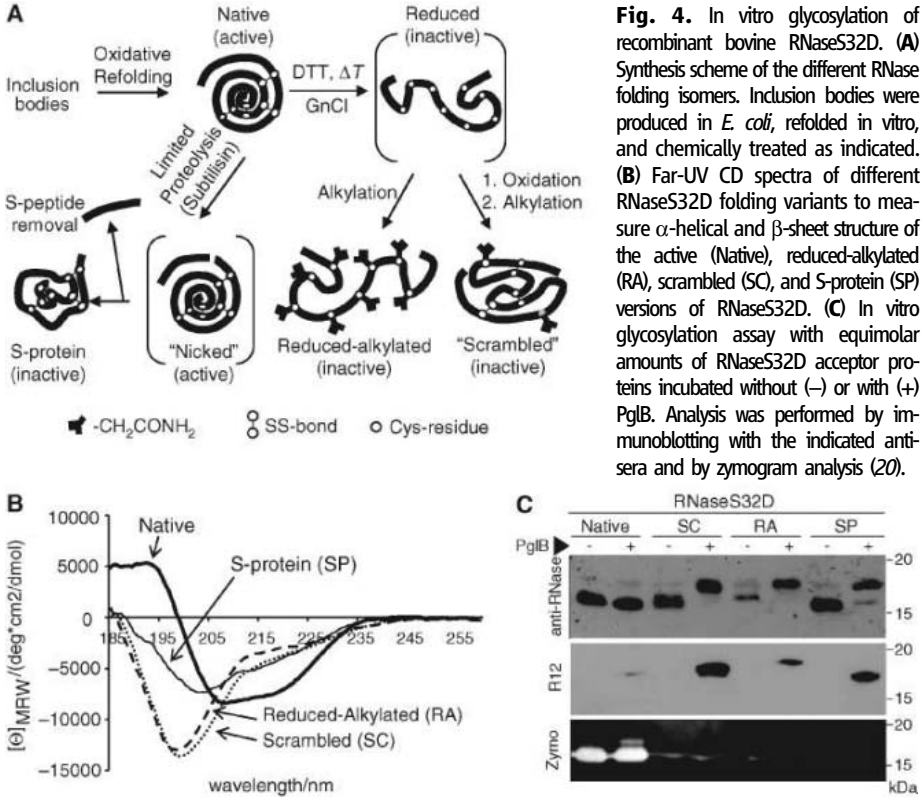

Fig. 4. In vitro glycosylation of recombinant bovine RNaseS32D. (**A**) Synthesis scheme of the different RNase folding isomers. Inclusion bodies were produced in *E. coli*, refolded in vitro, and chemically treated as indicated. (**B**) Far-UV CD spectra of different RNaseS32D folding variants to measure α-helical and β-sheet structure of the active (Native), reduced-alkylated (RA), scrambled (SC), and S-protein (SP) versions of RNaseS32D. (**C**) In vitro glycosylation assay with equimolar amounts of RNaseS32D acceptor proteins incubated without (–) or with (+) PglB. Analysis was performed by immunoblotting with the indicated antisera and by zymogram analysis (*20*).

product was similar to that of unglycosylated GFP (Fig. 3D), and thus the glycosylation reaction occurred on the folded GFP. Thus, PglB modified a peptide displayed on a folded protein, although it is likely that the grafted loop itself is relatively flexible.

To check the folding-dependent recognition of consensus sequons by PglB, we analyzed the PglB-dependent in vitro glycosylation of the eukaryotic glycoprotein bovine ribonuclease A (RNaseA). The native N-glycosylation site N34 of this protein is located in a structured domain. Different folding variants of the RNaseA allowed us to analyze the effect of unfolding on glycosylation. Consequently, RNaseA was expressed with a single point mutation (S32D), producing a bacterial consensus site at the native N-glycosylation site N34. Oxidative refolding of RNaseS32D from inclusion bodies (*20*) yielded enzymatically active protein (fig. S3), showing that RNaseS32D was able to fold into its native conformation despite the mutation.

Chemical treatments with denaturing, reducing, oxidizing, and alkylating agents yielded two RNaseS32D oxidation isomers (Fig. 4A). In reduced and alkylated RNaseS32D (RA), all four disulfide bonds were reduced in denaturing solution, and cysteines were alkylated to inhibit further disulfide bond formation. Rapid oxidation before alkylation led to another set of oxidation isomers, "scrambled" RNaseS32D (SC). Scrambled RNases contain randomly oxidized SS bonds, producing a heterogeneous population of proteins. A third RNaseS32D form was synthesized by limited proteolysis with subtilisin (*20*), which removes the N-terminal 21 amino acids of the protein. The resulting RNase S-protein (SP), like the RA and SC forms, was enzymatically inactive (fig. S3), but retained its native disulfide bonds and thus about 50% secondary structure, whereas the SC and RA forms appeared as random coils, as judged from far-ultraviolet circular dichroism (far-UV-CD) spectroscopy (Fig. 4B) (*23*). All four forms served as substrates for PglB (Fig. 4C). Glycosylation of the active RNaseS32D occurred with low efficiency (Fig. 4C), and the small amount of glycosylated RNaseS32D was active, as indicated by the zymogram assay (*20*) (Fig. 4C). The other forms were modified quantitatively (Fig. 4C). Thus, nonstructured protein domains are better substrates for PglB glycosylation than are folded ones. No glycosylation at all was observed with the same substrate proteins that lacked the S32D mutation (fig. S4B).

Our results show that completely folded proteins can be glycosylated both in vivo and in vitro. Bacterial OTase glycosylates native AcrA protein as well as an acceptor sequence grafted into the active GFP protein. In contrast, fully folded RNaseS32D was weakly glycosylated, whereas partial or complete unfolding strongly improved substrate activity.

The observation that the folding states of the acceptor protein affects glycosylation efficiency leads us to conclude that a specific substrate conformation must be adopted during the glycosylation process, most likely the Asn-turn (*24*). This makes potential acceptor sites present in a fixed environment suboptimal substrates for the bacterial OTase. We predict that native glycosylation sites in bacterial proteins will be located in locally flexible structures.

In contrast, the coupling of glycosylation and translocation in eukaryotes releases N-glycosylation from such structural constraints and, in combination with the less stringent primary sequence requirement, results in a more versatile and general glycosylation system.

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Materials and Methods
SOM Text
Figs. S1 to S4
Table S1
References

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New Strategies for the Elimination of Polio from India

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The feasibility of global polio eradication is being questioned as a result of continued transmission in a few localities that act as sources for outbreaks elsewhere. Perhaps the greatest challenge is in India, where transmission has persisted in Uttar Pradesh and Bihar despite high coverage with multiple doses of vaccine. We estimate key parameters governing the seasonal epidemics in these areas and show that high population density and poor sanitation cause persistence by not only facilitating transmission of poliovirus but also severely compromising the efficacy of the trivalent vaccine. We analyze strategies to counteract this and show that switching to monovalent vaccine may finally interrupt virus transmission.

The World Health Assembly committed to the global eradication of polio in 1988. Since then, the eradication initiative has achieved great successes, eliminating polio from the Americas, the Western Pacific, and Europe. However, in recent years the number of reported cases has increased after export of infection from the handful of remaining endemic countries. The difficulty in eliminating these last reservoirs of poliovirus transmission has led some to question the feasibility of eradication (*1*). Particularly worrying is the ongoing transmission in India, the source of half the world's reported paralytic cases over the past decade. Children in India have re-

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ceived many more doses of vaccine than children in other endemic countries through an intensive supplementary immunization program. Understanding the cause of polio persistence in India is a public health priority, both for the elimination efforts and as a proof of concept for the global eradication initiative. Potential explanations for persistence include gaps in routine and supplemental vaccine coverage (*2*), poor vaccine efficacy (*3*), and conditions highly favorable for the transmission of fecal-oral pathogens, including high population density and poor sanitation (*2*, *4*). Here, we examine these hypotheses formally, using detailed surveillance data from 96,421 cases of acute flaccid paralysis (AFP) collected since 1997.

The reproductive number $R(t)$ of an infection is the number of secondary infections that result from a single infectious individual in the population at time t (*5*). We estimated $R(t)$ for type 1 poliovirus transmission from the dates of onset of paralysis of laboratory-confirmed AFP cases and estimates of the incubation and infectious period (*6*). Although only ~1 in 200 cases of wild poliovirus infection result in paralysis (*7*, *8*), and not all of these cases are reported, this estimate of $R(t)$ is independent of the ratio of infections to reported paralytic cases.

The annual variation in the estimated $R(t)$ is particularly marked, with peak transmission about 150% greater than the annual average, compared, for example, with about 30% for measles in industrialized countries (*9*) (Fig. 1). Despite such strong seasonal forcing of transmission, sharp annual peaks in incidence are observed, rather than the more complex dynamics observed for some childhood infections (*10*). This is due to the long infectious period for fecal-orally transmitted polio. In contrast to Uttar Pradesh (UP) and Bihar, the estimated annual average $R(t)$ for poliovirus transmission outside these states has remained below one for the past 3 years, indicating that endemic transmission is no longer supported (Fig. 1C). This is confirmed by analysis of the genetic sequences of wild poliovirus isolates from AFP cases, with all genetic lineages circulating in India since 2003 derived from lineages circulating in UP in earlier years (*11*). The spatiotemporal dynamics of polio incidence now resemble classic "sink-source" dynamics (*12*), with the virus persisting in UP and Bihar (the "source") and expanding during the high-transmission season to infect other areas, including major cities such as Mumbai, but without the establishment of long-term transmission in these areas (Movie S1).

Why is poliovirus transmission persisting in UP and Bihar? Logistic regression reveals a significant association between continued reporting of laboratory-confirmed polio AFP cases by districts during 2000 to 2005 and (i) population density, (ii) the prevalence of diarrhea, and (iii) low routine coverage with three doses of trivalent oral polio vaccine (tOPV), after accounting for differences in the absolute number of children in each district (table S1). Those districts predicted by the regression model to have persistent poliovirus transmission are located mainly in UP and Bihar (Fig. 2). Of course, the regression simply reveals an association between these factors and continued reporting of poliovirus. This may be useful programmatically to identify those districts at higher risk of poliovirus transmission. However, there is likely to be a causal role for high population density, and the poor sanitary conditions that lead to a high prevalence of diarrhea, in the persistence of polio, consistent with the importance of these risk factors for other fecal-oral pathogens (*13*, *14*). High population density and poor sanitation can lead to more frequent infectious contacts and increase levels of excreted poliovirus in the environment. Routine immunization is also likely to be important, providing the very young with a dose of tOPV before they receive doses through supplementary immunization activities. However, routine immunization coverage relies on existing health services and is therefore confounded by socioeconomic and sanitary conditions. This is confirmed in the logistic regression, where the fraction of households reported to have a latrine was significantly associated with polio persistence when routine coverage was excluded from the analysis (table S2).

Children in India receive the majority of their tOPV doses through supplementary immunization activities. These have been increasingly focused on UP and Bihar, such that since 2004, children in these states were reported to have on average received more doses of vaccine than children in other parts of India (Fig. 1, D and E). In fact, at the end of 2005, children under 5 years old were reported to have received on average 15 doses of tOPV in UP and Bihar, compared with 10 in the rest of India, and only 4% of children were reported to have received fewer than 3 doses, of whom 90% were under 6 months old. Even under conditions highly favorable for the fecal-oral transmission of wild poliovirus, this level of vaccine coverage should have eliminated infection. We therefore estimated the efficacy of tOPV by comparing the reported number of doses of vaccine received by polio cases with nonpolio AFP controls (*6*). We found a decline in the relative odds of infection with paralytic polio with increasing number of doses of tOPV that is consistent with a constant, but unexpectedly low, probability of protection per dose (Fig. 3A). The estimated protective efficacy against type 1 poliovirus was just 9% per dose in UP, significantly lower than an estimated 21% per dose in the rest of India (Table 1). Similar results are obtained for

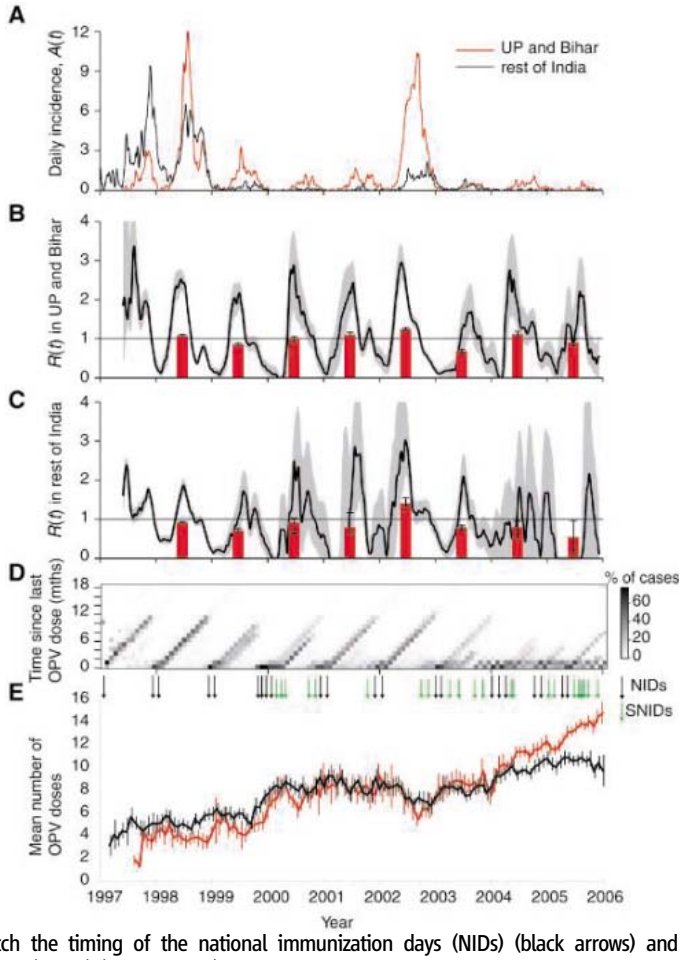

Fig. 1. Poliovirus and vaccination dynamics, 1997 to 2005. (**A**) The estimated daily incidence of laboratory-confirmed type 1 paralytic poliovirus $A(t)$ in UP and Bihar (red) and the rest of India (black). (**B** and **C**) The estimated reproductive number $R(t)$ in (B) UP and Bihar and (C) the rest of India, averaged over a sliding window of 43 days (the mean infectious period; solid line) and over a year (red bars). The gray areas and error bars present the bootstrap 95% confidence intervals about these estimates. (**D**) The distribution of reported time since last OPV dose and (**E**) average number of OPV doses received among 0- to 4-year-old children in UP and Bihar (red) and the rest of India (black), based on age-standardized nonpolio AFP cases. The "pulsed" declines in the average number of doses, and increases in time since last OPV dose, closely match the timing of the national immunization days (NIDs) (black arrows) and subnational immunization days (SNIDs) (green arrows).

type 3 poliovirus, although confidence intervals (CI) are wider, reflecting the lower number of reported cases. Estimates of vaccine efficacy were not significantly affected by the period of analysis or the year of onset of paralysis.

It is well known that trivalent OPV tends to be less efficacious in developing countries (even after accounting for vaccine formulation, quality, and administration) because of host and environmental problems, particularly interference with seroconversion by other enteroviruses and failure of the vaccine virus to establish infection in children with diarrhea (*15*–*17*). However, the per-dose estimate of efficacy for UP is significantly lower than earlier estimates of ~30%, based on seroconversion and small case-control studies in India (*15*, *18*–*20*) [as compared with ~65% in industrialized countries (*21*)]. Pre-release potency testing of tOPV used in India has been satisfactory and loss of potency before administration is unlikely, because vaccine is distributed rapidly and vaccine vial monitors have been used since 1998 to ensure that only vaccine stored at the right temperature is used. Vaccine efficacy may be underestimated if controls are less exposed to wild poliovirus than cases, or if parents overreport the number of doses of vaccine received by their children. However, the potential for differences in exposure was minimized by closely matching cases and controls by location, age, and date of onset of paralysis, and the matching criteria were chosen such that estimates of efficacy were robust to their value (*6*). Also, although some over-reporting of doses may occur, sensitivity analysis demonstrates that a vaccine efficacy comparable to the ~30% per dose found in earlier studies would require reporting of four doses for every one dose received, which is inconsistent with detailed case investigations. Instead, the lower efficacy in UP compared with earlier, mainly urban, studies is likely to be the result of more severe environmental problems. This conclusion is supported by the significantly lower efficacy estimated for tOPV administered in UP compared with other states where population is less dense and sanitary conditions are better.

High population densities and poor sanitation therefore appear to explain the persistence of polio. These factors act to facilitate the transmission not only of poliovirus but also of other enteroviruses and diarrhea, which interfere with the live-attenuated oral vaccine. Therefore, despite the higher number of doses received by children in UP and Bihar, we estimate that only 71% of children under 5 years old in these states were successfully immunized against type 1 poliovirus in early 2005, compared with 85% in the rest of India [based on vaccine coverage estimated from the nonpolio AFP data (*6*)].

The government of India and its partners have responded to this problem with a combination of new approaches and vaccine strategies. The currently high reported coverage with tOPV and low vaccine efficacy means that benefits from increasing vaccine efficacy will outweigh those from increasing vaccine distribution (Fig. 3B). For example, doubling the efficacy of the current vaccine would be equivalent to increasing the average number of doses received by children in UP and Bihar from 15 to 28. The global eradication of type 2, and the elimination of type 3 polio cases in recent years from all of India except a cluster of districts in western Uttar Pradesh, has motivated the introduction of monovalent vaccine, effective only against type 1 poliovirus, to immunization days in selected states beginning in April 2005. Monovalent vaccine has potentially higher efficacy than the trivalent vaccine because of the absence of interference with the two other OPV types (*22*). However, exclusive use of monovalent vaccine during immunization days can put the population at risk of outbreaks of type 3 poliovirus.

Mathematical analysis shows that the optimal balance of monovalent and trivalent vaccine use depends on the relative efficacy of the monovalent vaccine and the transmissibility (basic reproductive number R_0) of type 1 compared with type 3 wild poliovirus (Fig. 3C). Earlier studies of seroconversion from developing countries, including India, suggest a relative monovalent vaccine efficacy between 2.0 and 2.5 times that for trivalent vaccine (*15*, *22*, *23*). If types 1 and 3 were to have equivalent R_0, then support for monovalent vaccine use would be borderline (Fig. 3C). However, the lower incidence of type 3 despite broadly equivalent efficacy of tOPV against types 1 and 3—both in this study (Table 1) and in developing-country studies of seroconversion after administration of "balanced" formulations of tOPV (*15*)—suggests a lower transmissibility and/or lower pathogenicity (case-to-infection ratio) for type 3 (*8*). Lower transmissibility is consistent with the observation of a lower prevalence of antibodies against type 3 in India before vaccination (*24*, *25*). In this case, use of monovalent vaccine

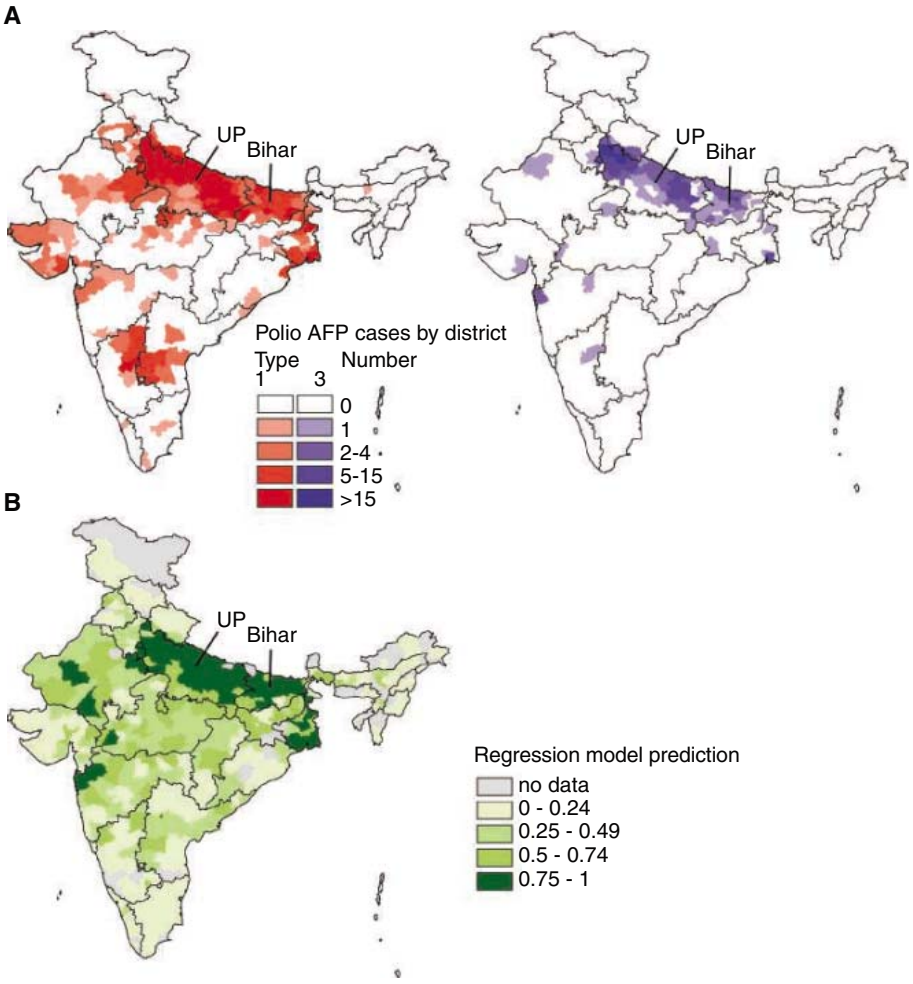

Fig. 2. Location of persistent poliovirus transmission in India. (**A**) Incident laboratory-confirmed type 1 and type 3 polio cases by district for the period 2000 to 2005. (**B**) Regression model estimate of the probability of poliovirus persistence (≥1 case of polio over 2000 to 2005) in each district, based on the number and density of children, the reported prevalence of diarrhea, and routine coverage with three doses of tOPV.

against type 1 is supported, with the amount distributed depending on the relative transmissibility (Fig. 3C). In districts where type 3 has been absent for several years, a greater fraction of vaccine doses distributed can be monovalent, depending on the risk of importation of type 3. In these districts, monovalent vaccine use has the potential to halve the population susceptible to type 1, assuming that coverage is maintained at its current level, substantially increasing the probability of interrupting transmission.

With new vaccine strategies based on careful use of monovalent vaccine targeted at districts with high population densities and poor sanitation, the analyses presented here suggest that wild poliovirus could soon be eliminated from India. Achieving this goal may also be facilitated by future improvements in sanitation, which can reduce transmission of both poliovirus and other infections that interfere with tOPV. Critical to the success of these new strategies will be continued dialogue and engagement with local communities to ensure high coverage with the appropriate vaccine.

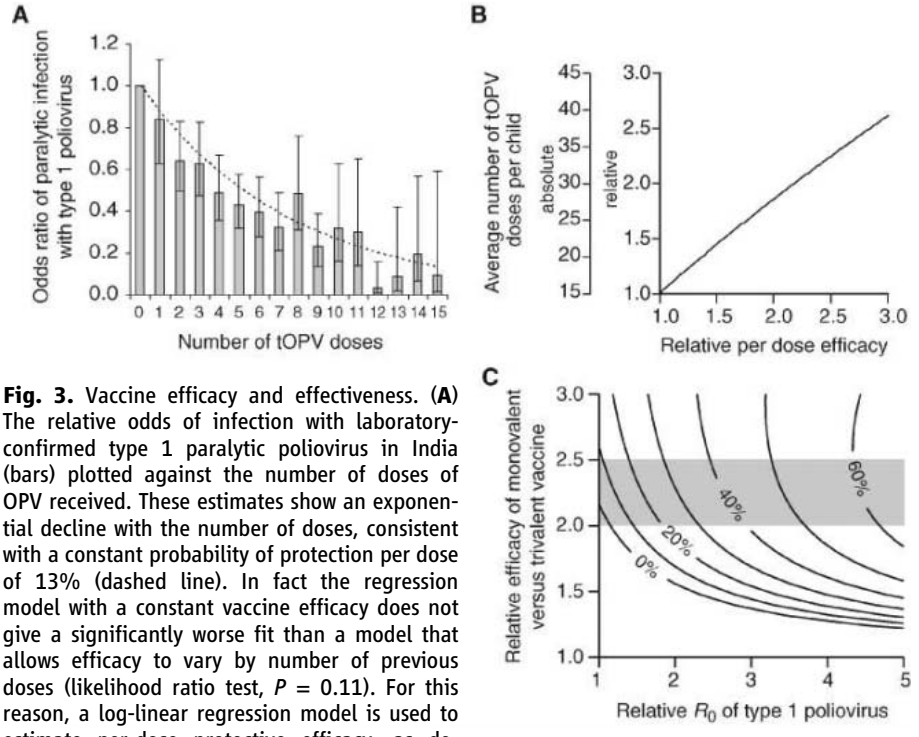

Fig. 3. Vaccine efficacy and effectiveness. (**A**) The relative odds of infection with laboratory-confirmed type 1 paralytic poliovirus in India (bars) plotted against the number of doses of OPV received. These estimates show an exponential decline with the number of doses, consistent with a constant probability of protection per dose of 13% (dashed line). In fact the regression model with a constant vaccine efficacy does not give a significantly worse fit than a model that allows efficacy to vary by number of previous doses (likelihood ratio test, $P = 0.11$). For this reason, a log-linear regression model is used to estimate per-dose protective efficacy, as described in (*6*). The error bars indicate 95% confidence intervals. (**B**) The absolute and relative increase in the average number of doses of OPV received by children less than 5 years old that would be required to achieve the same reduction in the effective reproductive number for poliovirus transmission as a given increase in vaccine efficacy. (**C**) A contour plot indicating the percentage of all doses of OPV administered that should be monovalent type 1 to minimize the average effective reproductive number of wild poliovirus types 1 and 3, for different relative efficacies of the monovalent versus trivalent vaccine and different relative transmissibility R_0 of type 1 versus type 3 (*6*). The expected relative efficacy of the monovalent vaccine is between 2 and 2.5 and is highlighted on the plot by the gray rectangle. Below the 0% contour, all doses should be trivalent. In (B) and (C), vaccine efficacy is assumed to be 9%, with an average child having received 15 doses of vaccine, in agreement with data from UP at the end of 2005.

Table 1. Estimates of trivalent OPV efficacy in India, 1997 to 2005. The per-dose protective efficacy of the vaccine was estimated from the reported number of OPV doses received by polio AFP cases compared with matched nonpolio AFP controls, using conditional logistic regression (*6*). Regression model 1 provides an estimate for all India, whereas model 2 includes an interaction term between efficacy and location.

Poliovirus	Regression model	Location	Cases	Matches	Vaccine efficacy (%) (95% CI)
Type 1	Model 1	All India	4421	1627	13 (10–16)
	Model 2	Rest of India	1512	361	21 (15–27)
		Bihar	387	158	18 (9–26)
		Uttar Pradesh	2522	1108	9 (6–13)*
Type 3	Model 1	All India	1204	474	13 (7–18)
	Model 2	Rest of India	221	79	21 (8–33)
		Bihar	136	53	22 (4–36)
		Uttar Pradesh	847	342	9 (3–15)

*Significantly different from rest of India, $P < 0.01$

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Supporting Online Material

www.sciencemag.org/cgi/content/full/314/5802/1150/DC1
Materials and Methods
Figs. S1 and S2
Tables S1 and S2
Movie S1

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The Psychological Consequences of Money

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Money has been said to change people's motivation (mainly for the better) and their behavior toward others (mainly for the worse). The results of nine experiments suggest that money brings about a self-sufficient orientation in which people prefer to be free of dependency and dependents. Reminders of money, relative to nonmoney reminders, led to reduced requests for help and reduced helpfulness toward others. Relative to participants primed with neutral concepts, participants primed with money preferred to play alone, work alone, and put more physical distance between themselves and a new acquaintance

People long have debated the effects of money on human behavior. Some scholars have pointed to its role as an incentive, insofar as people want money in order to trade it for prized goods or services (*1*, *2*). Others, however, have deplored money for undermining interpersonal harmony (*3*). We propose that both outcomes emerge from the same underlying process: Money makes people feel self-sufficient and behave accordingly.

In this Report, "money" refers to a distinct entity, a particular economic concept. Consistent with other scholarly uses of the term (*1*), we use the term money to represent the idea of money, not property or possessions. Our research activates the concept of money through the use of mental priming techniques, which heighten the accessibility of the idea of money but at a level below participants' conscious awareness. Thus, priming acts as a nonconscious reminder of the concept of money.

We tested whether activating the concept of money leads people to behave self-sufficiently, which we define as an insulated state wherein people put forth effort to attain personal goals and prefer to be separate from others. The term as we define it does not imply a value judgment and encompasses a mixture of desirable and undesirable qualities, which may help explain the positive and negative consequences of money (*4*).

The self-sufficiency hypothesis encapsulates findings from extant research on money. If money brings about a state of self-sufficiency, then a lack of money should make people feel ineffectual. Previous research indicates that physical and mental illness after financial strain due to job loss is statistically mediated by reduced feelings of personal control (*5*). A recent theory by Lea and Webley (*1*), which characterizes money as both a tool and a drug, emphasizes that people value money for its instrumentality: Money enables people to achieve goals without aid from others. Therefore, we predicted that reminders of money would lead to changes in behavior that suggest a feeling of self-sufficiency. When reminded of money, people would want to be free from dependency and would also prefer that others not depend on them.

In Experiment 1, participants were randomly assigned to three conditions. In two conditions (play money and money prime), participants were reminded of money; control participants were not reminded of money (*6*). All participants first completed a descrambling task (*7*), which activated neutral concepts (control and play money) or money (money prime). The descrambling task consisted of 30 sets of five jumbled words. Participants created sensible phrases using four of the five words. In the control and play-money conditions, the phrases primed neutral concepts (e.g., "cold it desk outside is" became "it is cold outside"). In the money-prime condition, 15 of the phrases primed the concept of money (e.g., "high a salary desk paying" became "a high-paying salary"), whereas the remaining 15 were neutral phrases (*6*). Participants in the play-money condition were primed with money by a stack of Monopoly money in their visual periphery while completing the neutral descrambling task.

Next, participants were given a difficult but solvable problem that involved arranging 12 disks into a square with five disks per side. As the experimenter exited the room, he offered that he was available to help if the participant wanted assistance. Persistence on the problem before asking for help was the dependent measure (*8*).

As predicted, participants who were reminded of money (play money and money prime) worked longer than control participants before requesting help [$F(2,49) = 3.73$, $P < 0.04$; mean (M) money prime = 314.06 s, SD = 172.79; M play money = 305.22 s, SD = 162.47; M control = 186.12 s, SD = 118.09]. The two money conditions did not differ from each other [$t(49) < 1$], but each was significantly different from the control group [money prime versus control: $t(49) = 2.44$, $P < 0.02$; Cohen's $d = 0.86$; play money versus control: $t(49) = 2.30$, $P < 0.03$; Cohen's $d = 0.84$]. Percentages of participants who requested help are shown in Fig. 1A.

In Experiment 2, we made two key changes to increase the generalizability of the findings of Experiment 1. First, we equated status differences between the would-be helper and the participant to ensure that differences in requests for help in Experiment 1 were not due to differential sensitivity to the experimenter's higher status. The second change was to the manipulation of the money prime. We hypothesized that money primes are unlikely to activate the idea of meager finances – rather, monetary wealth is probably what is activated. This reasoning suggests that directly reminding people of meager finances will not lead to the same effects as reminders of financial affluence, which we tested systematically in Experiment 2.

Participants were randomly assigned between two manipulations; one condition activated the idea of an abundance of money (high money) and the other activated the idea of restricted amount of money (low money). Participants first read aloud an essay in front of a video camera. Participants in the high-money condition read about growing up having abundant financial resources, whereas low-money participants read about growing up having meager resources. Afterward, all participants were given the opportunity to ask for help.

The indicator of self-sufficiency was persistence on an impossible task before asking for help. The participant's job was to outline all segments of a geometric figure once and only once without lifting the pencil or retracing any segments. Unbeknownst to participants, the figure was unsolvable. After 2 min of working alone, the experimenter and a confederate (who was blind to the participant's condition) entered the room. The experimenter said

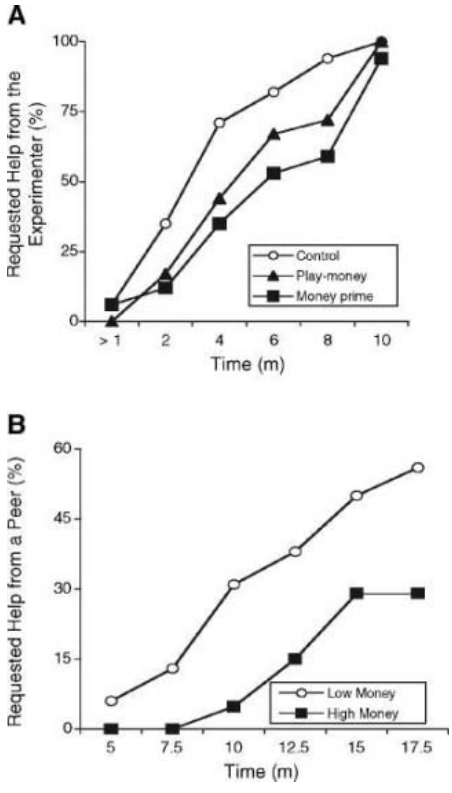

Fig. 1. Percentage of participants who asked for help as a function of money prime and length of time that had elapsed while working on (**A**) a difficult task (from Experiment 1) or (**B**) an unsolvable task (from Experiment 2).

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that the confederate was another participant who had just completed this experiment and therefore could be asked for help, if needed.

Results indicated that participants in the high-money condition worked significantly longer than participants in the low-money condition before asking for help [$t(35) = 2.03$, $P = 0.05$; Cohen's $d = 0.65$; M high money = 1058.48 s, SD = 210.12; M low money = 876.63 s, SD = 334.42]. Percentages of participants asking for help are shown in Fig. 1B. Thus, the effects of money did not depend on relative status differences between the participant and the helper.

In Experiment 3, we predicted that people who value self-sufficiency would be less helpful than others because they expect that each person will take care of him- or herself. Hence, we expected that participants primed with money would volunteer less time relative to control participants. Participants were randomly assigned to one of two conditions, one that primed money and one with neutral concepts. The priming manipulations were the money and neutral (control condition) descramble tasks from Experiment 1.

After the priming task, the experimenter explained that she was an undergraduate who was looking for help coding data and asked whether the participant would be able to help (*9*). She explained that each data sheet takes approximately 5 min to code. Participants were left alone to indicate how many data sheets, if any, they would be willing to code and also to provide their contact information.

Participants in the money condition volunteered to help code fewer data sheets than did participants in the control condition [$t(37) = 2.06$, $P < 0.05$; Cohen's $d = 0.66$] (Table 1). Translated into time, control condition participants volunteered an average of 42.5 min of their time, whereas participants in the money condition volunteered only slightly more than half that much (~25 min).

Experiment 3 showed that participants primed with money offered less help to the experimenter than did participants primed with neutral concepts. Yet, it may be that by asking for help for sometime in the future, the experimenter suggested that she was not in dire straits (in which case, she likely would have asked for immediate aid); thus, money condition participants may have failed to realize that help was truly needed. Accordingly, it was important to move beyond promises of help to measuring real helping behavior.

In Experiment 4, two between-subject conditions were used to prime money or neutral concepts. Each participant completed the descramble tasks (from Experiment 1). Next, the participant was left alone to complete irrelevant questionnaires. Meanwhile, the experimenter reentered with a confederate (who was blind to the participant's priming condition) and introduced her as another participant. The experimenter explained that there was no space in the laboratory and therefore the confederate must share a room with the participant. After pretending to work for one minute, the confederate asked the participant to explain the directions for the task she was given because she did not understand what to do. Time spent helping the confederate was the measure of helping.

Participants who were primed with money were less helpful than participants not primed with money [$t(42) = 2.13$, $P < 0.04$; Cohen's $d = 0.63$]. The data showed that participants primed with money spent half as much time helping the confused confederate as did participants in the control condition (Table 1). Apparently, participants who were primed with money believed that the confederate should figure out on her own how to perform the task, as a self-sufficient person would do.

In Experiment 5, we wanted to give money-primed participants a helping opportunity that required no skill or expertise, given that the help that was needed in the two previous experiments may have been perceived as requiring knowledge or special skill to enact. The opportunity to help in the current experiment was quite easy and obvious, in that it involved helping a person who spilled a box of pencils.

Participants were randomly assigned to one of three conditions that were manipulated in two steps. Each participant first played the board game Monopoly with a confederate (who was blind to the participant's condition) posing as another participant. After 7 min, the game was cleared except for differing amounts of play money. Participants in the high-money condition were left with $4000, which is a large amount of Monopoly money. Participants in the low-money condition were left with $200. Control condition participants were left with no money. For high- and low-money participants, the play money remained in view for the second part of the manipulation. At this step, participants were asked to imagine a future with abundant finances (high money), with strained finances (low money), or their plans for tomorrow (control).

Next, a staged accident provided the opportunity to help. A new confederate (who was blind to the participant's priming condition) walked across the laboratory holding a folder of papers and a box of pencils, and spilled the pencils in front of the participant. The number of pencils picked up (out of 27 total) was the measure of helpfulness.

As predicted, the money prime influenced helpfulness [$F(2, 32) = 4.34$, $P < 0.03$]. Participants in the high-money condition gathered fewer pencils than did participants in the low-money condition [$t(32) = 2.75$, $P < 0.02$; Cohen's $d = 0.81$] or those in the control condition [$t(32) = 2.13$, $P < 0.05$; Cohen's $d = 1.23$] (Table 1). Helpfulness did not differ between the low-money group and the control group [$t < 1$, not significant). Even though gathering pencils was an action that all participants could perform, participants reminded of financial wealth were unhelpful.

Experiment 6 tested for the psychological effects of money by operationalizing helpfulness as monetary donations. Upon arrival to the laboratory, participants were given $2 in quarters in exchange for their participation. The quarters were said to have been used in an experiment that was now complete; in actuality, giving participants quarters ensured that they had money to donate (*9*).

Participants were randomly assigned to one of two conditions, in which they descrambled phrases (as in Experiment 1) that primed money or neutral concepts. Then participants completed some filler questionnaires, after which the experimenter told them that the experiment was finished and gave them a false debriefing. This step was done so that participants would not connect the donation opportunity to the experiment. As the experimenter exited the room, she mentioned that the lab was taking donations for the University Student Fund and that there was a box by the door if the participant wished to donate. Amount of money donated was the measure of helping. We found that Participants primed with money donated significantly less money to the student fund than participants not primed with money [$t(38) = 2.13$, $P < 0.05$; Cohen's $d = 0.64$] (Table 1).

To convincingly demonstrate that money makes people self-sufficient, we tested the hypothesis in new contexts. The final experiments tested the effects of money on social intimacy, desire to engage in leisure activities alone, and preference to work alone. In Experiment 7,

Table 1. Helpfulness as a function of experimental condition in Experiments (Exp.) 3 to 6. The data are means ± SD; higher numbers indicate greater helpfulness. Within each experiment, means from the money and no-money conditions are different from each other at $P < 0.05$.

Exp. no.	Money condition	No-money condition	Dependent variable
3	5.10 ± 3.99	8.47 ± 5.99	Number of data sheets participants volunteered to code
4	67.35 ± 84.65	147.81 ± 158.15	Time spent helping a peer (seconds)
5	18.00 ± 1.96	20.30 ± 1.77 (control) 19.72 ± 2.28 (low money)	Number of pencils gathered
6	0.77 ± 0.74	1.34 ± 1.02	Monetary donations (in $)

participants were randomly assigned to one of three priming conditions. Participants sat in front of a computer while completing questionnaires. After 6 min, one of three screensavers appeared. Participants in the money condition saw a screensaver depicting various denominations of currency floating underwater (fig. S1). Participants in the fish condition saw a screensaver with fish swimming underwater (fig. S2). Participants in the no-screensaver condition saw a blank screen.

Afterwards, participants were told they would be having a get-acquainted conversation with another participant. Participants were asked to move two chairs together while the experimenter left to retrieve the other participant. The dependent measure was distance between the two chairs (*10*).

Participants primed with money placed the two chairs farther apart than did participants in the fish condition [$t(33) = 2.37$, $P < 0.05$; Cohen's $d = 1.07$] and the no-screensaver condition [$t(33) = 2.30$, $P < 0.05$; Cohen's $d = 0.85$] (Table 2). Chair distance did not differ between fish and blank screensaver conditions [$t(33) < 1$, not significant]. Hence, participants primed with money put more physical distance between themselves and a new acquaintance than participants not primed with money.

In Experiment 8, we tested whether money-primed participants would place a premium on being alone even when choosing leisure activities that could be enjoyed with friends and family. Participants were randomly assigned to one of three priming conditions. Participants first sat at a desk, which faced one of three posters, to complete filler questionnaires. In the money condition, the desk faced a poster showing a photograph of various denominations of currency (fig. S3). In two control conditions, the desk faced a poster showing either a seascape or a flower garden (figs. S4 and S5).

Subsequently, participants were presented with a nine-item questionnaire that asked them to choose between two activities. Within each item, one option was an experience that only one person could enjoy and the other option was for two people or more (e.g., an in-home catered dinner for four versus four personal cooking lessons).

Participants primed with money chose more individually focused leisure experiences than participants primed with either of the two neutral primes [$F(2, 58) = 4.04$, $P < 0.05$; money versus seascape: $t(58) = 2.75$, $P < 0.05$; Cohen's $d = 0.59$; money versus flowers: $t(58) = 2.10$, $P < 0.05$; Cohen's $d = 1.06$] (Table 2). The choice of activities did not differ between neutral conditions [$t(58) < 1$, not significant]. Thus, money primes lead people to be less social relative to those in nonmoney prime conditions.

In Experiment 9, a more rigorous test of the self-sufficiency hypothesis was tested: We asked whether people reminded of money would choose to work alone. Working on a task with a co-worker presumably means less work for each person, but the co-worker may prefer to rely on the participant, which would be an affront to self-sufficiency. Participants were given the option of working on a project with a peer or alone. Participants were randomly assigned to three priming conditions. As in Experiment 7, screensavers showing money, fish, or no screensaver primed money or nonmoney concepts. Participants were then told that their next task was an advertisement development task on which they could work alone or with a peer. Participants were left alone to indicate their choice.

Participants' desire to work with a peer was significantly affected by priming condition [$X^2(2, n = 37) = 10.10$, $P < 0.01$] (Table 2). Choosing to perform the task with a co-worker was reduced among money condition participants relative to participants in both the fish [$X^2(1) = 7.00$, $P < 0.05$; odds ratio = 11.25] and no-screensaver conditions [$X^2(1) = 8.22$, $P < 0.05$; odds ratio = 15.00]. There was no difference in choice between the fish and no-screensaver conditions [$t(34) < 1$, $P > 0.05$, not significant].

Nine experiments provided support for the hypothesis that money brings about a state of self-sufficiency. Relative to people not reminded of money, people reminded of money reliably performed independent but socially insensitive actions. The magnitude (*11*) of these effects is notable and somewhat surprising, given that our participants were highly familiar with money (*12*) and that our manipulations were minor environmental changes or small tasks for participants to complete.

Research on the repercussions of studying economics dovetails nicely with our results. Frank, Gilovich, and Regan (*13*) reported that university students majoring in economics made self-interested moves in social dilemma games more often than students of other disciplines. Economics students also were more convinced than noneconomists that their competitors would make self-interested moves, a result that echoes the present thesis that money evokes a view that everyone fends for him- or herself.

The self-sufficient pattern helps explain why people view money as both the greatest good and evil. As countries and cultures developed, money may have allowed people to acquire goods and services that enabled the pursuit of cherished goals, which in turn diminished reliance on friends and family. In this way, money enhanced individualism but diminished communal motivations, an effect that is still apparent in people's responses to money today.

Table 2. Social distance preferences as a function of experimental condition in Experiments (Exp.) 7 to 9. The data are means ± SD; higher numbers indicate preferences for greater social distance. In Experiments 7 and 9, the neutral 1 condition represents the fish screensaver condition, whereas the neutral 2 condition represents the no-screensaver condition. In Experiment 8, the neutral 1 condition represents the flower poster, whereas the neutral 2 condition represents the seascape poster. Within each experiment, means for the money condition differ from means in both neutral conditions at $P < 0.05$.

Exp. no.	Money condition	Neutral 1 condition	Neutral 2 condition	Dependent variable
7	118.44 ± 41.63	79.48 ± 30.43	80.54 ± 47.06	Physical distance between participant and partner (centimeters)
8	4.00 ± 1.20	2.82 ± 1.00	3.10 ± 1.80	Number of solitary activity selections
9	0.83 ± 0.39	0.31 ± 0.48	0.25 ± 0.45	Proportion of participants who opted to work alone

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Supporting Online Material

www.sciencemag.org/cgi/content/full/314/5802/1154/DC1
Materials and Methods
Figs. S1 to S5
References

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Generation of Gut-Homing IgA-Secreting B Cells by Intestinal Dendritic Cells

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Normal intestinal mucosa contains abundant immunoglobulin A (IgA)–secreting cells, which are generated from B cells in gut-associated lymphoid tissues (GALT). We show that dendritic cells (DC) from GALT induce T cell–independent expression of IgA and gut-homing receptors on B cells. GALT-DC–derived retinoic acid (RA) alone conferred gut tropism but could not promote IgA secretion. However, RA potently synergized with GALT-DC–derived interleukin-6 (IL-6) or IL-5 to induce IgA secretion. Consequently, mice deficient in the RA precursor vitamin A lacked IgA-secreting cells in the small intestine. Thus, GALT-DC shape mucosal immunity by modulating B cell migration and effector activity through synergistically acting mediators.

The gut harbors the large majority of IgA-secreting cells found in the body. After activation of naive B cells by the B cell receptor (BCR), newly generated IgA-producing cells leave the GALT, enter the blood, and home to the intestinal lamina propria, where they are required for optimal protection against intestinal pathogens (*1*, *2*). However, BCR stimulation in nonmucosal tissues produces few gut-homing IgA-secreting cells, which suggests that specific mucosa-associated differentiation signals induce both class switching to IgA and targeting of the ensuing effector B cells to the gut. Previous studies have shown that gut-homing T cells acquire their tissue tropism in response to imprinting signals from GALT-DC (*3*, *4*). Moreover, recent findings indicate that DC can present unprocessed antigens to B cells (*5*, *6*) and influence the differentiation and survival of antibody-secreting cells (ASC) (*7*). Here, we asked whether DC might contribute to the acquisition of tissue-specific functional properties of B cells, particularly their migration and/or effector activity in the gut.

Lymphocytes that infiltrate the gut mucosa display a distinct set of adhesion molecules (*2*). In particular, gut-tropic T cells and ASC express the integrin α4β7 and the chemokine receptor CCR9, which are essential for lymphocyte migration to the small intestine (*2*). GALT-DC selectively induce these traffic molecules in activated T cells (*3*, *4*) because GALT-DC, unlike DC from other lymphoid tissues, synthesize RA. The presence of this vitamin A metabolite is sufficient to induce gut-homing receptors on activated T cells, even in the absence of GALT-DC (*8*).

Given the impact of GALT-DC–derived RA on effector T cell migration, we hypothesized that this mechanism might also target B cells to the gut. Indeed, addition of DC from Peyer's patches (PP-DC) or RA to activated murine spleen B cells induced high levels of α4β7 and maintained robust CCR9 expression on B cells, whereas pooled DC from inguinal, axillary, and brachial peripheral lymph nodes (PLN-DC) or RA-free media induced $\alpha4\beta7^{-/low}CCR9^{low}$ B cells (Fig. 1, A to D, and figs. S1 and S2). Using optimized culture conditions (SOM Text 1) that improved the yield of viable B cells (figs. S3, A and B, and S4, A to C), we then performed in vivo homing experiments. As expected, RA substantially boosted B cell migration to the small bowel (Fig. 1, E and F).

Next, we asked whether human GALT-DC operate similarly. Naive (IgD^+CD27^-) and antigen-experienced (IgD^-CD27^+) human spleen B cells were activated with antibody to IgM together with autologous DC from human spleens, livers, or mesenteric lymph nodes (MLN-DC). Similar to the murine system, autologous DC also improved the yield of viable human B cells (figs. S3, C and D, and fig. S4D), and activated B cells that were exposed to liver- or spleen-DC expressed much less α4β7 and CCR9 than B cells that were activated together with MLN-DC or liver/spleen-DC plus RA (Fig. 2, A and B). Imprinting of gut tropism by MLN-DC depended on RA, because α4β7 and CCR9 induction was abrogated by LE540, which blocks the RAR family of RA receptors (*9*). Thus, the cellular and molecular mechanisms that imprint gut-homing B cells are conserved across species.

Gut tropism was not acquired only by naive human B cells; $CD27^+$ antigen-experienced B cells, initially $\alpha4\beta7^{-/low}CCR9^{-/low}$, also became $\alpha4\beta7^{high}CCR9^+$ upon activation in the presence of RA or MLN-DC (Fig. 2, A and B), suggesting that memory B cells retain migratory plasticity, analogously to memory T cells (*10*, *11*). To explore this more rigorously, we stimulated B cells with or without RA and then restimulated under reversed imprinting conditions. B cells that were activated without RA expressed few gut-homing receptors but became $\alpha4\beta7^{high}CCR9^+$ upon restimulation with RA (Fig. 2, C and D). Conversely, B cells first exposed to RA and then restimulated without RA down-regulated CCR9 but remained $\alpha4\beta7^{High}$ (Fig. 2C and SOM Text 2). Thus, effector B cells adjust their homing commitment to changing microenvironmental conditions. Accordingly, in repeatedly immunized humans the ASC homing phenotype depends on the route of antigen entry during the most recent immunization, irrespective of previous immunizations (*12*).

In vivo, human and murine IgA-ASC express the chemokine receptor CCR10, which has been implicated in ASC migration to mucosal tissues (*13*). However, neither RA nor PP-DC induced B cell responsiveness to CCR10 agonists, even though B cells migrated toward the CCR9 ligand CCL25/TECK (fig. S1B and SOM Text 3).

Having established that GALT-DC–derived RA induces gut-tropic B cells, we asked whether GALT-DC also induce IgA secretion, the hallmark effector activity of mucosal B cells. Indeed, activated B cells cultured with PP-DC secreted much more IgA than B cells activated without DC or with PLN-DC (Fig. 3A and fig. S5). This activity required only PP-DC but not T cell help or other environmental factors. Previous studies had reported induction of IgA secretion by B cells cocultured with PP-DC, but only in the presence of T cells (*14*). However, T cell–independent IgA induction has been observed in vivo (*15*), but the factors that induce IgA-ASC in this setting were not identified.

Because only PP-DC, but not PLN-DC, induced IgA-ASC, we asked whether this tissue-specific difference was due to differential cytokine production. In particular, IL-6 induces IgA responses in vivo and in vitro (*14*, *16*). However, adding antibody to IL-6 to B cell/PP-DC cocultures had only a modest, nonsignificant effect on IgA production (Fig. 3B). IL-5 is also implicated in IgA responses (*17*), but antibody to IL-5 did not decrease PP-DC–driven IgA production (Fig. 3B).

Because no single candidate cytokine was essential for PP-DC–induced IgA-ASC generation, we hypothesized that RA might be involved. This prediction seemed reasonable because the addition of RA to LPS-activated splenocytes promotes IgA secretion (*18*). Moreover, vitamin A deficiency impairs intestinal IgA responses, and vitamin A supplementation restores IgA levels in malnourished mice (*19*, *20*). Indeed, RA receptor blockade by LE540 decreased IgA levels in B cell cocultures with PP-DC (Fig. 3B). Concomitant inhibition of both RA and IL-6 had an additive effect, suggesting that these agents cooperate for optimal IgA induction. These observations were reproducible with VI10/Yen B cells expressing a switchable transgenic vesicular stomatitis virus (VSV)–specific BCR; exposure of bona fide naive VI10/Yen B cells to ultraviolet-

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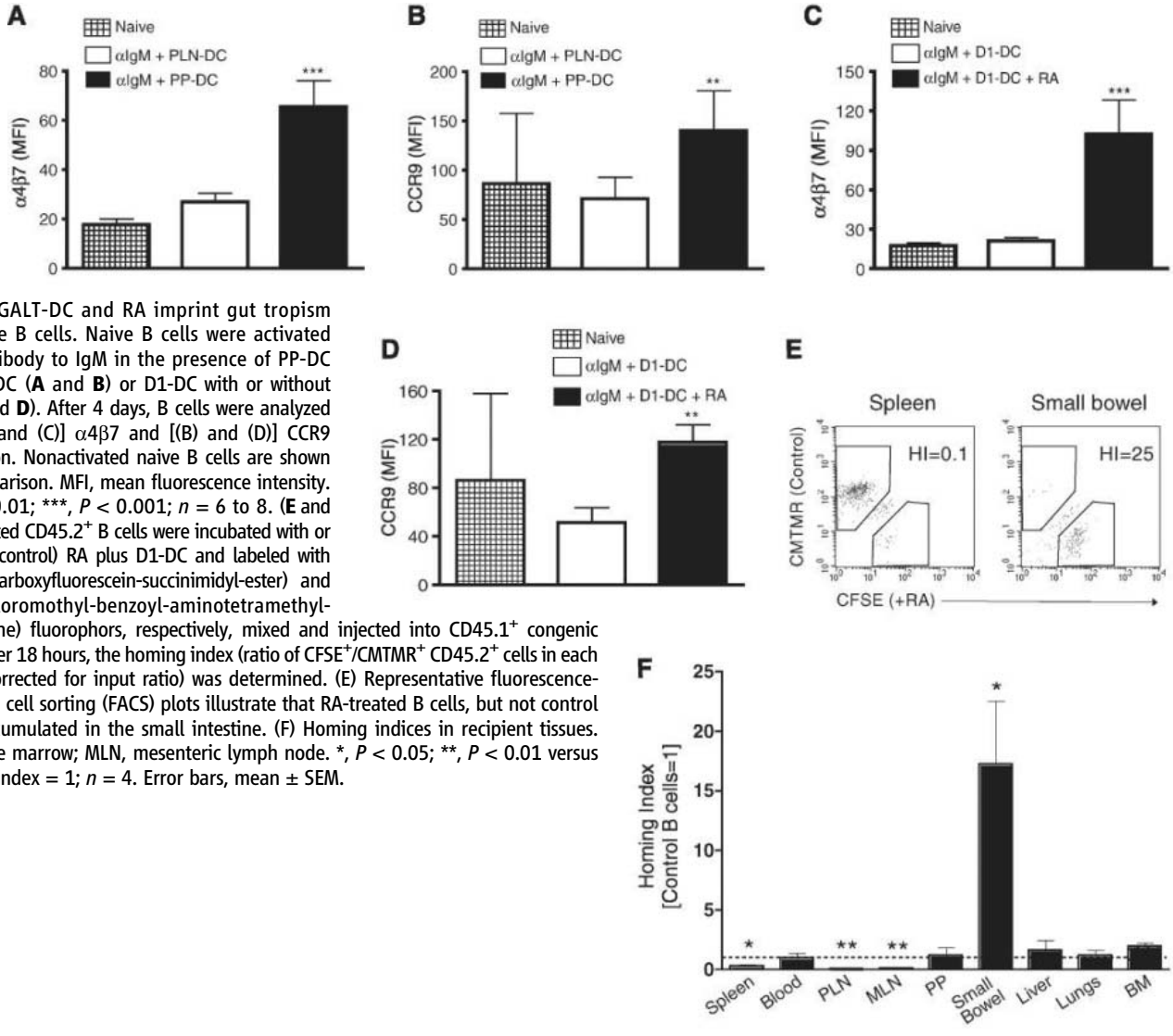

Fig. 1. GALT-DC and RA imprint gut tropism in mouse B cells. Naive B cells were activated with antibody to IgM in the presence of PP-DC or PLN-DC (**A** and **B**) or D1-DC with or without RA (**C** and **D**). After 4 days, B cells were analyzed for [(A) and (C)] $\alpha4\beta7$ and [(B) and (D)] CCR9 expression. Nonactivated naive B cells are shown for comparison. MFI, mean fluorescence intensity. **, $P < 0.01$; ***, $P < 0.001$; $n = 6$ to 8. (**E** and **F**) Activated CD45.2$^+$ B cells were incubated with or without (control) RA plus D1-DC and labeled with green (carboxyfluorescein-succinimidyl-ester) and red (chloromothyl-benzoyl-aminotetramethyl-rhodamine) fluorophors, respectively, mixed and injected into CD45.1$^+$ congenic mice. After 18 hours, the homing index (ratio of CFSE$^+$/CMTMR$^+$ CD45.2$^+$ cells in each tissue, corrected for input ratio) was determined. (E) Representative fluorescence-activated cell sorting (FACS) plots illustrate that RA-treated B cells, but not control cells, accumulated in the small intestine. (F) Homing indices in recipient tissues. BM, bone marrow; MLN, mesenteric lymph node. *, $P < 0.05$; **, $P < 0.01$ versus homing index = 1; $n = 4$. Error bars, mean ± SEM.

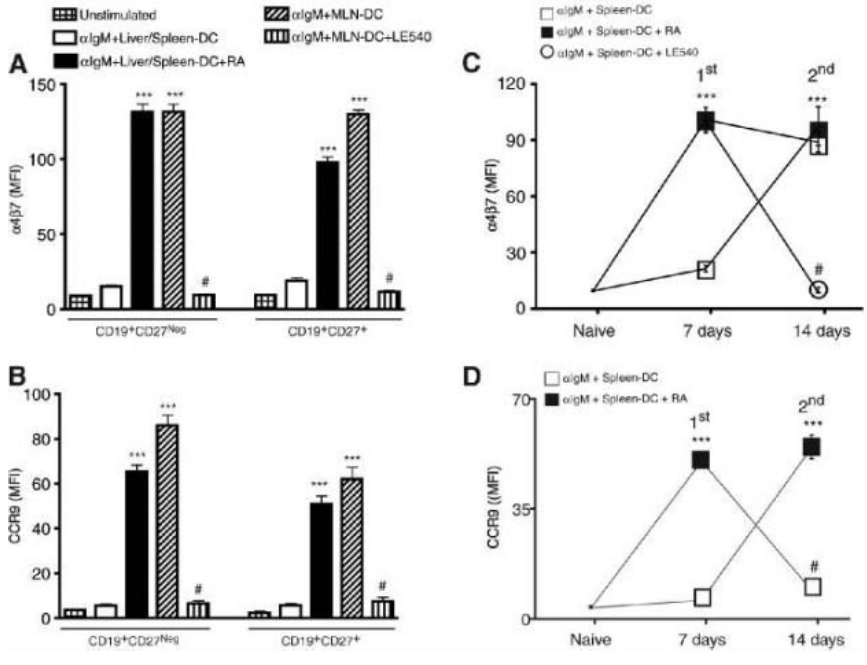

Fig. 2. Gut-homing receptor regulation on human B cells. Human spleen–derived B cells (95% CD19$^+$) from six donors were separated into CD27$^-$ (naive) and CD27$^+$ (antigen-experienced) subsets. (**A** and **B**) Cells were stimulated with antibody to IgM in the presence of liver-DC or spleen-DC with or without RA (100 nM), or with MLN-DC with or without LE540 (1 μM). After 7 days, CD19$^+$ cells were analyzed for (A) $\alpha4\beta7$ and (B) CCR9 expression. ***, $P < 0.001$ versus naive B cells or B cells plus liver/spleen-DC; #, $P < 0.001$ versus MLN-DC. (**C** and **D**) Naive B cells from three donors were cultured with spleen-DC and antibody to IgM with or without RA (1st stimulation). On day 7, B cells were analyzed for (C) $\alpha4\beta7$ and (D) CCR9, reactivated with or without RA or LE540 (2nd stimulation), and reanalyzed on day 14. ***, $P < 0.001$ versus concomitant $\alpha4\beta7^{Low}$ or $CCR9^{Low}$ sample; #, $P < 0.001$ versus spleen-DC. Error bars, mean ± SEM.

inactivated VSV plus PP-DC elicited notable IgA secretion (fig. S6), indicating that PP-DC induced de novo IgA class switching rather than expansion and/or survival of already committed IgA-ASC.

To determine whether RA plus IL-5 and/or IL-6 are sufficient to induce IgA secretion, we stimulated B cells with or without PLN-DC and different combinations of RA and cytokines (Fig. 3C and fig. S7). Each factor by itself or together with PLN-DC induced negligible IgA production, and combined IL-5 plus IL-6 was also ineffective. However, RA plus IL-5 and/or IL-6 and PLN-DC substantially enhanced IgA production. This synergistic effect of RA plus IL-5/IL-6 with PLN-DC was much more pronounced than the effect of B cell coculture with PP-DC. Indeed, PP-DC–induced IgA production was also boosted by RA plus IL-5/IL-6 (Fig. 3D and SOM Text 4). Given that IL-5 and IL-6 synergized with RA but not with each other, their effect on IgA secretion might require a shared signaling pathway, possibly involving JAK2 and STAT3 (*21*, *22*). Although this apparent redundancy of IL-5 and IL-6 (and possibly other cytokines) awaits further in vivo exploration, this effect could explain why genetic deficiency in IL-6 does not abrogate antigen-specific IgA responses (*23*). Transforming growth factor–β (TGF-β) has also been implicated in IgA switching/secretion (*24*, *25*), but TGF-β1 did not induce IgA secretion in this setting, even in combination with RA and/or IL-5/IL-6 (fig. S8 and SOM Text 5).

Interestingly, RA plus IL-5/IL-6 did not induce IgA-ASC in the absence of DC, which suggests that DC contribute additional essential factor(s). This effect was observed not only with murine D1-DC, PP-DC, and PLN-DC but also with human DC from liver, spleen, and MLN (Fig. 3E, fig. S7, and SOM Text 6). Human MLN-DC, but not liver-DC or spleen-DC also possessed a potent intrinsic capacity to promote human IgA-ASC (Fig. 3E). Human MLN-DC–driven IgA secretion depended on DC-derived RA and IL-6, and supplementation of RA plus IL-6 to human B cells augmented IgA secretion in cultures containing liver-DC, but not in the absence of DC.

Having established that GALT-DC–derived RA is essential to induce gut-homing IgA-ASC in vitro, we investigated the physiological role of RA using vitamin A–deficient mice, in which GALT-DC lack the prerequisite substrate for RA production (*8*). Compared with control mice, most secondary lymphoid organs in vitamin A–deficient animals contained fewer $\alpha4\beta7^+$ antigen-experienced (i.e., $B220^+/IgD^-$) B cells (Fig. 4A). By contrast, the frequency of all $B220^+/IgD^-$ cells (which include memory B cells) in vitamin A–deficient mice was decreased only in Peyer's patches but was normal or increased

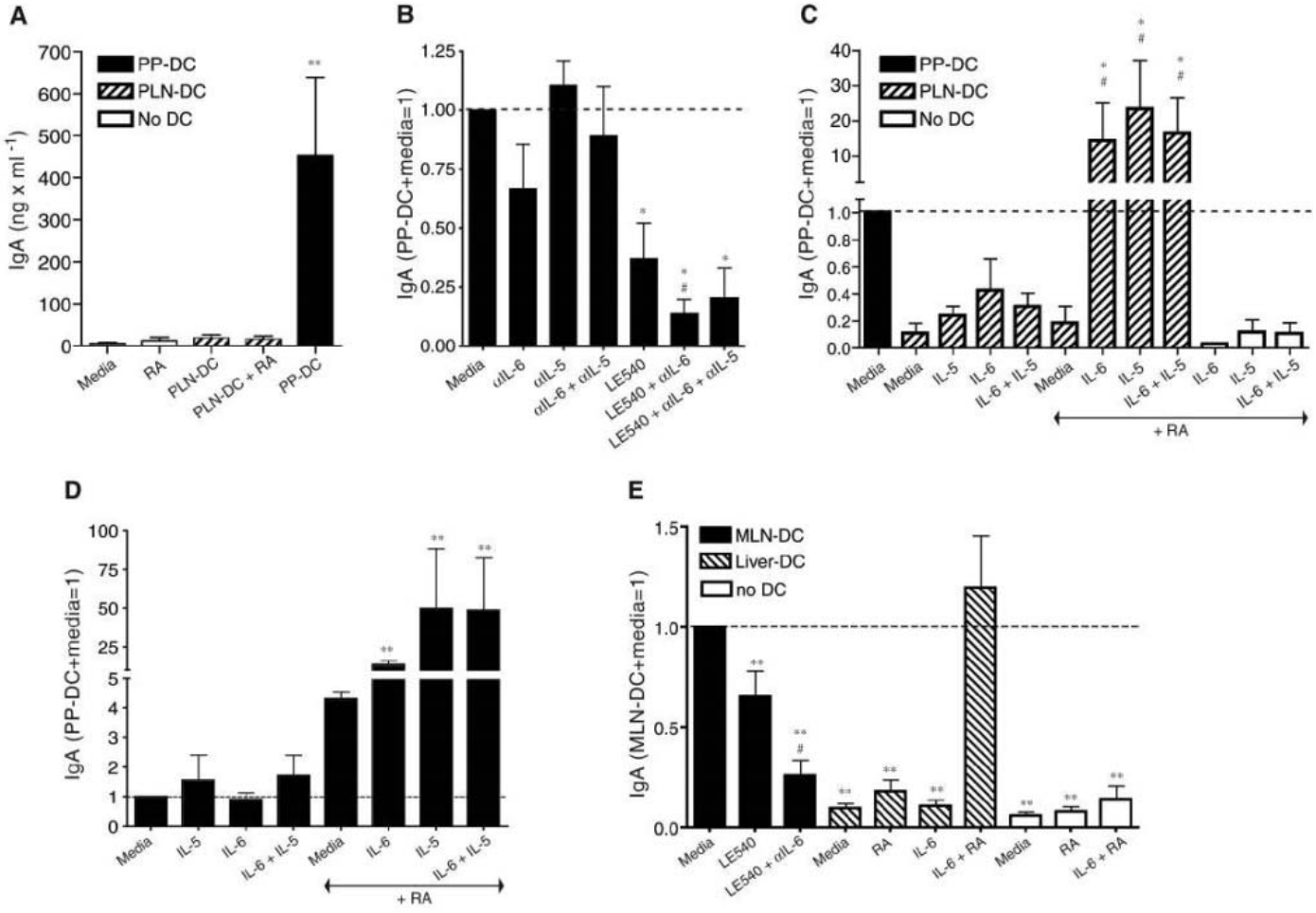

Fig. 3. Synergistic effects of RA and cytokines induce IgA-ASC. (**A**) Activated mouse B cells were cocultured with PP-DC or PLN-DC with or without RA. IgA levels in supernatants were determined by enzyme-linked immunosorbent assay after 4 days. **, $P < 0.01$ versus all groups; $n = 8$. (**B**) Activated mouse B cells were cultured with PP-DC and antibody to IL-5 and/or antibody to IL-6 (10 μg/ml) with or without LE540 (1 μM). *, $P < 0.01$ versus media; #, $P < 0.05$ versus antibody to IL-6; $n = 4$. (**C**) Naive mouse B cells were activated alone or in DC cocultures with or without exposure to IL-5 and/or IL-6 (10 ng/ml) and/or RA. *, $P < 0.05$ versus PP-DC and PLN-DC+media; #, $P < 0.05$ versus respective samples without RA; $n = 3$ to 8. (**D**) Activated mouse B cell cocultures with PP-DC were treated with cytokines and/or RA. *, $P < 0.05$ versus media+RA; $n = 3$ to 4. (**E**) Human B cells were activated with antibody to IgM alone or with human liver-DC or MLN-DC and indicated reagents. After 7 days, IgA levels in supernatant were measured. **, $P < 0.01$ versus media; #, $P < 0.05$ versus MLN-DC+LE540; $n = 3$ to 6. Error bars, mean ± SEM.

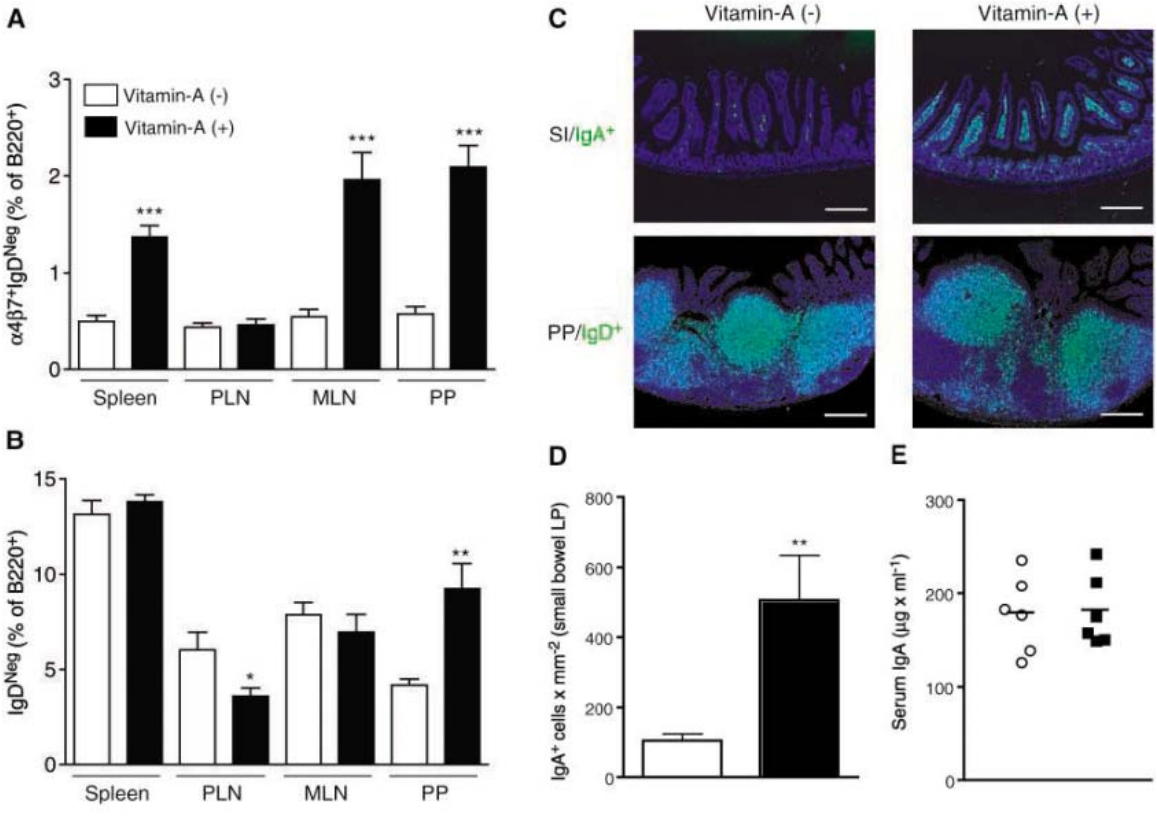

Fig. 4. Role of dietary vitamin A in the generation of gut-homing B cells and intestinal IgA-ASC. Vitamin A–deficient (–) and –sufficient (+) mice were generated as described in (*29*). The frequency of $\alpha4\beta7^{+}IgD^{-}$ $B220^{+}$ cells (**A**) and total IgD^{-} $B220^{+}$ cells (**B**) in lymphoid organs was determined by FACS; $n = 8$ mice per group. (**C**) Immunohistochemistry illustrates the paucity of IgA^{+} cells in the small intestine [(SI), upper row)] of vitamin A–deficient mice (left) compared with control mice (right). Naive IgD^{+} B cells were normal in Peyer's patches (PP, lower row) irrespective of vitamin A status. Scale bar, 200 μm. (**D**) Quantitative histological analysis of IgA^{+} cell numbers in intestinal lamina propria; $n = 5$ sections from 2 mice per group. (**E**) Serum IgA levels; $n = 6$. In (A), (B), and (E), vitamin A–sufficient and vitamin A–deficient mice were compared using a two-tailed unpaired Student's *t* test, whereas results in (D) were compared by the Mann-Whitney U test. *, $P < 0.05$;**, $P < 0.01$;***, $P < 0.001$. Error bars, mean ± SEM.

in other lymphoid organs (Fig. 4B). Thus, vitamin A deficiency does not compromise antigen-experienced B cell numbers per se but affects preferentially the intestinal effector/memory repertoire. Indeed, as reported previously in rats (*26*), IgA^{+} cells were substantially reduced in the lamina propria of vitamin A–deficient mice (Fig. 4, C and D). Vitamin A deficiency also affected IgA^{+} cell frequencies in GALT and spleen (figs. S9 and S10), suggesting that the paucity of intestinal IgA-ASC was caused not only by the inability of GALT-resident B cells to acquire gut-homing receptors but also by defective differentiation of IgA-ASC in lymphoid tissues.

Despite the lack of intestinal IgA-ASC, vitamin A–deficient mice had normal serum IgA levels (Fig. 4E), indicating that IgA-ASC do not obligatorily depend on RA. Extraintestinal IgA-ASC express traffic molecules distinct from gut-homing receptors (*2*), whose induction is probably RA-independent.

We conclude that GALT-DC induce gut tropism and IgA secretion in B cells by overlapping, yet distinct, mechanisms. The characteristic ability of GALT-DC to synthesize RA is sufficient to imprint gut tropism. However, to generate IgA-ASC, RA must synergize with cytokines produced by the DC and/or other cells. Although there are probably alternative mechanisms generating extraintestinal IgA-ASC, RA appears critical for IgA-ASC to populate the small bowel. The clinical relevance of this tissue-restricted mechanism is underscored by the observation that vitamin A supplementation decreases the incidence and severity of diarrhea in malnourished children (*27*), resulting in mortality reduction by as much as 34% (*28*).

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Medical Biotechnology Center UMBI

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Diversity—Women in Science

Building Better Balance

Women leaders in science—from a university president and policy makers to an academic researcher and industrial scientists—see improvements in this field's gender balance, but they also know that more must be done. An increasing number of women are entering scientific studies in college, but better ways are needed to keep these women in science and to help them grow into leadership positions. BY MIKE MAY

When it comes to women in science these days, their success shows just how much they have accomplished. Nonetheless, more work remains to put men and women on equal ground in science and engineering. "There's good news and bad news, " says Shirley M. Malcom, head of the American Association for the Advancement of Science's directorate for education and human resource programs. "Women have flocked to the biological sciences, and we nearly have parity with men at the doctoral level," she says. Clearly, that is good news. On the other hand, says Malcom, "There is not the advancement expected, given the participation by women in science."

In September, the U.S. National Academies released a new report called "Beyond Bias and Barriers: Fulfilling the Potential of Women in Academic Science and Engineering" (available at http://www.nap.edu/catalog/11741.html). This report indicates that women earn more than half of all bachelor's degrees in science and engineering, and almost 40 percent of the doctoral degrees in the same areas. Nonetheless, fewer women than men make the transition to faculty careers, and those women who do "typically receive fewer resources and less support than their male colleagues," according to this report. In part, this shows that attrition impacts women much more than men along the path from science student to science professional. "There's leakage of women in every step of the system," says Donna E. Shalala, president of the University of Miami and chair of the panel that wrote the report. Still, she says, "This is an enormously positive report." That upbeat perspective reflects a crucial success: Today's leaders know how the system must be refined to bring in and retain more women in this area in the future. CONTINUED »

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Diversity—Women in Science


DONNA J. DEAN

A Review of Role Models

For young women to aspire to careers in science and engineering, they need high-powered leaders to emulate. In academics and private organizations, there are several examples: including Susan Hockfield, president at the Massachusetts Institute of Technology; Shirley Ann Jackson, president of Rensselaer Polytechnic Institute; Judith Rodin, president of the Rockefeller Foundation; and Shirley Tilghman, president of Princeton University. Women also hold high-level positions in science organizations run by the government. For example, Donna J. Dean, president of the Association for Women in Science and senior science adviser at Lewis-Burke Associates, LLC, points out that at the U.S. National Institutes of Health four of 27 institute directors are women: Patricia Grady, director of the National Institute of Nursing Research; Story C. Landis, director of the National Institute of Neurological Disorders and Stroke; Nora D. Volkow, director of the National Institute on Drug Abuse; and Elizabeth G. Nabel, director of the National Heart, Lung, and Blood Institute. In scientific and engineering industries, women can also be found in leadership roles: including Stephanie Burns, chairman and chief executive officer of Dow Corning; Susan Desmond-Hellmann, president of product development at Genentech; and Una Ryan, chief executive officer at AVANT Immunotherapeutics.

Nonetheless, even successful women in science see the field as male dominated. "I'm not aware of any area in academics, government, or industry in which many women hold top positions," says Cris Lewis, head of biochemical pharmacology in small-molecule drug discovery at Genentech. "Women are still clearly in the minority." Still, opportunities do exist for women. "While not all women aspire to management and upper level positions," says Janice Kameir, vice president of human resources at Diversa, "the opportunities are there if women take responsibility for their own career success." She adds, "Throughout industry, there are women in management roles in scientific research and business, but we need to actively find ways to support and encourage women who want to move ahead in our industry."

In part, having role models can encourage young women to enter and stick with science. Lesley Murray, associate director of translational oncology at Genentech, says, "My path may have been somewhat unusual, since I had mostly female supervisors and mentors, and have also had the privilege of knowing many inspiring women in very senior positions as strong role models." That experience gives Murray a positive outlook. She says, "Opportunities for women to rise to the top clearly exist in industry, and I have personally never encountered any prejudice because of my gender."

Young women interested in careers in science or engineering would also benefit from seeing more female role models covered in the media. This year, one pair of women scientists—Elizabeth H. Blackburn of the University of California, San Francisco, and Carol W. Greider of the Johns Hopkins University School of Medicine—gained considerable publicity as winners of the 2006 Albert Lasker Award for Basic Medical Research. That kind of recognition helps aspiring women scientists and engineers think positive and think big.


JANICE KAMEIR

A Call to Computing

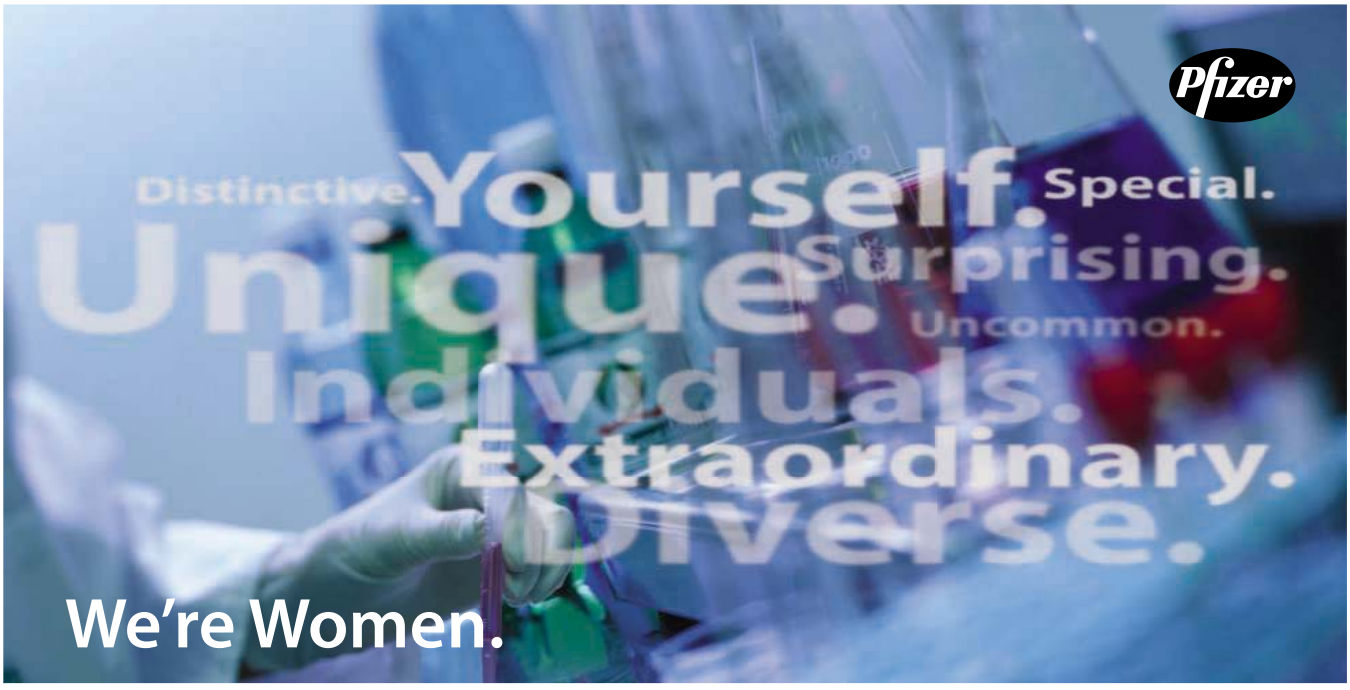
Some areas—including computer science—look very appealing for women in the future. Cynthia Breazeal, associate professor of media arts and sciences at the Massachusetts Institute of Technology's Media Lab, says, "This is an incredibly rich area careerwise." She believes that young students might bypass this field because they think it's just about sitting down and programming. "In principle," Breazeal says, "this field can be applied to many different disciplines. As in any field, computer science is a constellation of techniques and theories and methodologies. It is much broader than simply learning C++ or Java."

To encourage students to take up computing, Breazeal believes that introductory courses must take a new approach. Students should be shown the ties between programming and exciting fields, including artificial intelligence, multimedia applications, robotics, and so on. "We need to make it engaging, tangible, and more exciting," says Breazeal. In part, that means getting students into labs where they can see computing in action. "This field hasn't homed in on the best presentation yet," adds Breazeal.

Overall, engineering complex systems demands a wide range of skills. For example, Breazeal says, "Robotics is extremely multidisciplinary—art, design, and technology. You must be more of a Renaissance thinker, so to speak." Breazeal got excited about robotics when she stumbled over "the deep interaction between technology and the life sciences." She wants to create an entity whose cognitive and affective processes are inspired by those that animals and people use when they behave in complex environments.

Institutional Evolution

Despite all of the advances for women in science, some serious obstacles must still be surmounted. "The more elite the organization," says Shalala, "the less likely you are to find women, whether you are discussing the National Academies, the National Institutes of Health, or full professors at research universities." In the past, a lack of qualified women candidates supposedly explained that inequity. "Now, the pools have changed," Shalala says. "There are no excuses anymore." CONTINUED »



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Diversity—Women in Science

Nonetheless, "Beyond Bias and Barriers" points out that a collection of unconscious actions creates a culture that does not support young women. For instance, Shalala says that the university system does not accommodate child-bearing among women faculty. Moreover, she says, "Academics has been a male culture, and a young female professor might not fit in as comfortably."

Malcom also sees problems in the current system. "There are things that institutions must do," she says. "Women need to be given the same front-end resources as males to go into high-end positions." Dean agrees. She says, "There are necessary cultural changes. For that, I would cite 'Beyond Bias and Barriers.' It is absolutely fantastic at laying out all of the systemic changes and approaches that organizations and institutions should take."

Some changes are already under way. At Princeton, for example, male and female faculty members get an extra year to work toward tenure if they have or adopt a child during the usual tenuring period.

Beneficial changes are also taking place in industry. Lewis sees great value in "institutions or companies, like Genentech, that offer policies and benefits that make the life of a working parent just a little bit easier every day, such as on-site child care, concierge services, financial, legal, dental, and automotive service, emergency rides for commuting parents with sick children, et cetera." She adds, "This can provide an extra edge that is a huge benefit for working mothers, so they don't feel that they are forced to choose between being a parent and still having a highly successful career."

MICHAEL COLELLA

SHIRLEY M. MALCOM

Better Molding

To help women work their way to upper end positions, Lewis says, "I believe mentoring can play a very valuable role. Supervisors in upper management positions can make a difference by providing straightforward feedback to ensure that women understand what skills or talents may need additional development." Other women in science agree with the value of mentoring. As Lewis says, "Sometimes, we need to be reminded how just a little bit of time can have a big influence on a young person's life." To learn more about mentoring, see the Association for Women in Science's *A Hand Up* (http://www.awis.org/pubs/ahandup.html) and MentorNet (http://www.mentornet.net).

Nonetheless, institutions and mentors cannot do all of the work. People must work to improve their odds of getting hired and advancing. "Those who hold top positions understand the value of developing professional competencies, such as management skills and strategic thinking," says Kameir. "Women who learn to master these skills—while leveraging their own scientific background—are in a good position to take on leadership roles." In addition, Kameir recommends a career plan "with specific action steps for achieving short-term and long-term goals." She says that this should include the development of strategic relationships. "If women don't understand the importance of strategic relationship building, they will work twice as hard for half the reward and recognition," Kameir says.

LESLEY MURRAY

In addition, women can even climb up by improving their own expectations. Murray says, "I would encourage women to work on their self-esteem and self-confidence, where we certainly lag behind many male colleagues." She adds, "Women scientists must learn how to be more assertive, without being overly aggressive, and make the time for networking with both male and female colleagues."

Also, advancement takes time and persistence. "There are a lot of skills that higher level positions call for that you build up over time," says Malcom. "You must be able to present well, be clear, be able to communicate in all kinds of spheres—written or oral." She adds that everyone can benefit from a knowledge of strategic planning, managing people, developing budgets, and understanding the role of policy development.

To develop a network and build skills, Murray encourages all female scientists to join the Association of Women in Science. Likewise, several of the other experts offered the same advice. Murray says, "I have been a member in Palo Alto, California, for several years, and have been impressed by the encouragement this organization gives—helping high school science students, awarding scholarships to community college students with hardships, and being a voice in Washington, D.C., on national issues of critical importance to women in science."

DONNA E. SHALALA

Getting a Head Start

In the long run, the scientific community could focus on women at even earlier ages. "It is a really crucial issue to get students excited in science and technology before the undergraduate level," says Breazeal. "How do we attract more girls from kindergarten through high school?" She has some ideas. "You can imagine an online game that could engage kids in doing science and scientific thinking in ways they care about, and they could have fun with it." But she adds, "It has to be participatory and engaging, and they have to care about it."

In the end, being successful depends on loving the work. "You might get discouraged some days," says Dean, "but if you love it, you will pursue it." Still, a career in science challenges anyone. "Nothing is perfect," says Dean. "Even though it can be hard—like encountering someone who says, 'Girls can't do science'—just say, 'I *can* do it!'"

Mike May (mikemay@mindspring.com) is a publishing consultant for science and technology based in the state of Minnesota, U.S.A.

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Graduate Student Fellowships

Iowa State University's faculty are recognized nationally and internationally for expertise in the biological sciences including genetics, bioinformatics, toxicology, food science and technology, genomics, biorenewable resources, and biomedical sciences. Graduate student applicants are eligible for research assistantships and fellowships. Full tuition scholarships are available for doctoral students. **http://www.grad-college.iastate.edu/programs/APprograms.php**

Prestigious Fellowships include: George Washington Carver Doctoral Fellowships; USDA National Needs Fellowships in Animal Genomics and Bioinformatics; USDA National Needs Fellowship in Food Safety and Quality; NSF IGERT Traineeships in Computational Molecular Biology; Alliance for Graduate Education and the Professoriate (AGEP); ISU Plant Sciences Institute (PSI) Fellowships. NIH and NSF Biomedical Research Assistantships are available.

http://www.genetics.iastate.edu/fellowship.html

Summer Undergraduate Research Opportunities

Are available in genetics; toxicology; bioinformatics and computational biology; nutrition; food science; animal, microbial and plant sciences; chemistry; molecular biotechnology; genomics; STEM fields: science, technology, engineering, and mathematics; statistics; emerging interface technologies and human computer interaction; bioeconomy; biological materials and processes; mathematics and computing; computation materials chemistry; natural resources; physics and in other areas.

http://www.genetics.iastate.edu/summerinternships.html

Iowa State University is located in Ames, Iowa, a community noted nationally for its exceptional quality of life.

Women and minorities are strongly encouraged to apply.

UNCF • MERCK SCIENCE INITIATIVE

"A mind is a terrible thing to waste"

MERCK

UNDERGRADUATE SCIENCE RESEARCH SCHOLARSHIP AWARDS

- 15 Awards Annually
- Scholarships up to $25,000
- Two Summer Internships at a Merck Research Facility

An applicant must:

- Be a full-time student at any four-year college or university
- Have junior year academic status
- Major in a life or physical science (first professional degrees excluded)
- Have a minimum cumulative GPA of 3.3 (4.0 point scale)

GRADUATE SCIENCE RESEARCH DISSERTATION FELLOWSHIPS

- 12 Fellowships Annually
- Fellowship Stipends up to $42,000
- Department Grants of $10,000
- Support for 12-24 months

An applicant must:

- Be enrolled full-time in a Ph.D. or equivalent doctoral program in a biomedical life or physical science
- Be engaged in and within 1-3 years of completing dissertation research

POSTDOCTORAL SCIENCE RESEARCH FELLOWSHIPS

- 10 Fellowships Annually
- Fellowship Stipends up to $70,000
- Department Grants of $15,000
- Support for 12-24 months

An applicant must:

- Hold a Ph.D. or equivalent degree in a biomedical life or physical science
- Be appointed as a new or continuing postdoctoral fellow by the end of 2007 at an academic or non-academic research institution (private industrial laboratories are excluded)

Applicants must be African American (Black), U.S. citizens or permanent residents, and attending an institution in the U.S.A. Applications must be submitted online at www.uncf.org/merck/ or postmarked by December 15, 2006

For more information, please contact your department chairperson or Jerry L. Bryant, Ph.D., at the **United Negro College Fund**, Inc., 8260 Willow Oaks Corporate Drive, P.O. Box 10444, Fairfax, VA 22031-4511, by fax (703) 205-3574, by e-mail at ***uncfmerck@uncf.org***.

NORTHWESTERN
UNIVERSITY

The greatness of a university is directly proportional to the quality of its intellectual community. People from similar backgrounds can and do learn from each other. However, new approaches to knowledge are most likely to be discovered when scholars from a wide variety of backgrounds are brought together to interact with and challenge one another. In a community like ours, social diversity is a mainspring of intellectual and creative progress and contributes directly to academic excellence….
From the 2001-02 Report of the Faculty Diversity Committee

SCIENCE AND ENGINEERING DEPARTMENTS CONDUCTING SEARCHES INCLUDE:

***Weinberg College of Arts and Sciences*: Biochemistry, Molecular Biology, and Cell Biology; Chemistry; Mathematics; Neurobiology and Physiology; Physics and Astronomy**

***McCormick School of Engineering and Applied Science*: Biomedical Engineering; Chemical and Biological Engineering; Civil and Environmental Engineering; Electrical Engineering and Computer Science; Engineering Science and Applied Mathematics; Materials Science and Engineering; Mechanical Engineering**

***Feinberg School of Medicine*: Microbiology—Immunology**

For details regarding these searches, please see:
www.northwestern.edu/provost/sci_eng_searches

Northwestern University is committed to further diversifying its faculty. We warmly encourage applications from women and members of under-represented minority groups.

CHAIRMAN, Department of Cell Biology University of Oklahoma Health Sciences Center

Applications/nominations are invited for Chair of the Department of Cell Biology at the University of Oklahoma Health Sciences Center. This individual must be committed to the Department's mission of biomedical research in cellular, developmental, and molecular biology, as well as medical education. The successful candidate will have an M.D., Ph.D., or equivalent doctoral degree, qualify for tenured appointment as Professor, and be an internationally recognized leader in his/her area of research. Candidates with interests in cancer biology and diabetes are particularly sought, given the Department's participation in the ongoing expansion of the OU Cancer Institute and the Oklahoma Diabetes Center. Leadership positions in the OUCI are available for qualified applicants. With over $180 million in public and private support, OUCI represents the largest investment in biomedical research in the state's history. Please send a letter of application, curriculum vitae, the names and contact information for five references, plus a one-page summary that includes teaching philosophy and goals for maintaining and expanding the Department to: **Dr. Robert Foreman, Chair, Cell Biology Chair Search Committee, The University of Oklahoma, BMSB 653, 940 Stanton L. Young Blvd., Oklahoma City, OK, 73104 (http://w3.ouhsc.edu/cell%5Fbiology/)**. The review of applications will begin immediately and continue until the position is filled.

The University of Oklahoma is an Equal Opportunity Institution.

Neurobiology of Disease Gladstone/UCSF Faculty Position

The Gladstone Institute of Neurological Disease and the University of California, San Francisco (UCSF) invite applications for a faculty position at the level of Assistant, Associate, or Full Professor. Of particular interest are neuroscientists and neurologists with broad expertise in chemistry, bioinformatics, or physiology who are interested in investigating experimental models of neurodegenerative disorders at the molecular, cellular, or systems level. Primary criteria for appointment will be outstanding records of innovative research and academic performance, including landmark papers in leading journals, as well as high potential for establishing a rigorous and substantial independent research program, inspiring mentorship, and fruitful collaboration.

The successful candidate will join an interactive group of investigators in Gladstone's state-of-the-art research facility at UCSF's new Mission Bay campus. S/he will have joint appointments in the Gladstone Institute of Neurological Disease, the Department of Neurology, the Neuroscience Graduate Program and the Biomedical Sciences Graduate Program at UCSF. Excellent salary support is provided. Women are especially encouraged to apply. The search will continue until the position is filled. To ensure full consideration, however, applications should be received by **December 31, 2006**. For additional information on research programs and facilities, see **www.gladstone.ucsf.edu/gind**.

Please send curriculum vitae, description of achievements and research interests, and the names of three references to:

GIND/UCSF Search Committee
1650 Owens Street
San Francisco, CA 94158
gindsearch@gladstone.ucsf.edu

The J. David Gladstone Institutes and UCSF are Affirmative Action/Equal Opportunity Employers. Gladstone and the University undertake affirmative action to assure equal employment opportunity for underutilized minorities and women, for persons with disabilities, and for Vietnam-era veterans and special disabled veterans. We seek candidates whose experience, teaching, research, or community service has prepared them to contribute to our commitment to diversity and excellence.

UCSF
University of California
San Francisco

Sandler Asthma Basic Research Center Assistant Professor

The Sandler Asthma Basic Research (SABRE) Center at the University of California San Francisco seeks outstanding applicants to join an expanding research unit dedicated to basic investigations of the pathogenesis of asthma. While grounded in basic molecular, cellular and biochemical studies, scientists in the SABRE Center interact closely with colleagues at UCSF that span the research spectrum from basic scientific pursuits to clinical and genetic translational studies of human asthma. Applicants must have M.D./Ph.D., Ph.D., or M.D. training, with demonstrated research creativity. Particular areas of interest include innate immunity, inflammation, dendritic cell biology, memory and mucosal defenses. Prior research in asthma is not required. A generous start-up package and access to core facilities in flow cytometry, genomics, microscopy, small animal physiology and genetics will assist scientists in setting up a research program in asthma. Applicants will be appointed as Assistant Professor in the appropriate Department and will participate in competitive graduate school programs at UCSF. Candidates with clinical training qualifying for appointments in the Departments of Medicine, Pediatrics or Pathology are encouraged to apply.

Candidates should send curriculum vitae, a two-page summary of research plans and the names and addresses of five references by **December 31, 2006** to:

Richard M. Locksley, M.D.
Director, SABRE Center
University of California, San Francisco
513 Parnassus Avenue, Rm. S-1032B
Campus Box 0795
San Francisco, CA 94143-0795

UCSF is an Affirmative Action/Equal Opportunity Employer. The University undertakes affirmative action to assure equal employment opportunity for under-represented minorities and women, for persons with disabilities, and for Vietnam-era veterans and special disabled veterans.

DIVERSITY - AWARDS

DORIS DUKE
CHARITABLE FOUNDATION

Announcing the 2007 Clinical Scientist Development Award Competition

Clinical Scientist Development Awards

Fostering Careers in Research

CALL FOR NOMINATIONS

This program offers three-year awards providing $135,000/year to help physician-scientists make the critical transition from training to independence as clinical investigators.

Nominations of clinical investigators at the instructor or assistant professor level are being solicited from accredited, degree-granting institutions in the United States. Institutions may nominate up to three candidates in any disease area. *Nominations of women and under-represented minorities in medicine are strongly encouraged.*

Full details and instructions are available at: www.ddcf.org/mrp-csda.

Application Deadlines

Nominations Due: January 23, 2007
Full Proposals Due: March 6, 2007
Award Start Date: August 1, 2007

DORIS DUKE
CHARITABLE FOUNDATION

Congratulations to the 2006 Clinical Scientist Development Award Recipients

Visit www.ddcf.org/mrp and join the Medical Research Program's listserv to receive announcements of upcoming competitions

Ferhaan Ahmad, M.D., Ph.D., University of Pittsburgh
Alessandro Cataliotti, M.D., Ph.D., Mayo Clinic
Michael K. Cooper, M.D., Vanderbilt University Medical Center
Grant Dorsey, M.D., Ph.D., University of California, San Francisco
Gianpietro Dotti, M.D., Baylor College of Medicine
Matthew L. Freedman, M.D., Dana-Farber Cancer Institute
Mardi Gomberg-Maitland, M.D., M.Sc., University of Chicago
Rosandra Kaplan, M.D., Weill Medical College of Cornell University
Ross L. Levine, M.D., Dana-Farber Cancer Institute
Rita Nanda, M.D., University of Chicago
Sanjiv M. Narayan, M.D., M.Sc., University of California, San Diego
Christopher Newton-Cheh, M.D., M.P.H., Massachusetts General Hospital
Anju Nohria, M.D., Brigham and Women's Hospital
W. Kimryn Rathmell, M.D., Ph.D., University of North Carolina, Chapel Hill
Harmony R. Reynolds, M.D., New York University School of Medicine
Kimberly A. Risma, M.D., Ph.D., Cincinnati Children's Hospital Medical Center
Michael B. Rothberg, M.D., M.P.H., Tufts University Medical Center
Manish Sagar, M.D., Brigham and Women's Hospital
Catherine S. Todd, M.D., M.P.H., University of California, San Diego
Thomas D. Wang, M.D., Ph.D., Stanford University
Jonathan Weinsaft, M.D., M.A., Weill Medical College of Cornell University
Mitchell David Wong, M.D., Ph.D., University of California, Los Angeles
Xu Yu, M.D., M.Sc., Massachusetts General Hospital

POSITIONS OPEN

CHAIR POSITION
Department of Pharmaceutical Sciences
Washington State University

A department chair position at the tenured full professor level is available to begin on or about September 1, 2007. Exceptional associate professor candidates will be considered. Candidates must have a Ph.D. and/or M.D. in a biomedical or related discipline and an active funded research program. Research in all areas will be considered, although pharmaceutical sciences, biomedical/bioengineering, molecular therapeutics and related fields are preferred. The successful candidate will have leadership capabilities and a commitment to expanding the research and teaching environment at WSU. Additional junior faculty positions are available to further grow the department. Demonstrated competence in the development and/or implementation of professional and graduate curricula and an excellent record of mentoring is desirable. The position may include an endowed professorship. See **http://www.pharmacy.wsu.edu/PharmSci/** for additional information. Screening will begin **January 1, 2007**. To apply, please submit a letter of application *explaining how your record relates to the qualifications and responsibilities listed*; a CV; and the names and contact information of 3 references. Send (e-mail acceptable) to: **Raymond M. Quock, Ph.D., College of Pharmacy, P.O. Box 646534, Washington State University, Pullman, WA 99164-6534, quockr@mail.wsu.edu**.

WSU is an EO/AA Educator and Employer.

NEUROETHOLOGY

The Section of Integrative Biology of the University of Texas at Austin seeks applications for an Assistant Professor in the area of **Neuroethology** to begin in September 2007. The successful applicant will join a strong program in behavior, ecology, and evolution, with strengths in sensory ecology, behavioral ecology, neuroendocrinology, behavioral genomics, phylogenetics, and population biology. Applicants may work on any organisms, but candidates working on arthropod neuroethology are particularly encouraged to apply. A Ph.D. is required in Biological Sciences or related areas and postdoctoral experience is preferred. Teaching duties will include an undergraduate course in animal behavior and a graduate course in the candidates' area of interest. The successful candidate will also be eligible for affiliation with the Institute of Neuroscience, which provides state-of-the-art core facilities and graduate program support (see **www.utexas.edu/neuroscience**).

Applicants should send a curriculum vitae, brief statements of research and teaching interests, up to 5 reprints/preprints, and arrange for three letters of recommendation. Application material should be sent as a single PDF file (including cover letter, vita, statements, and reprints/preprints) to: **ibjob@uts.cc.utexas.edu**. Letters of recommendation should be sent by regular mail to: **Neuroethology Search, Integrative Biology, 1 University Station C0930, Austin, TX 78712**. Review of applications will begin **2 January 2007**. For more detailed information see **http://www.biosci.utexas.edu/jobs/**.

UT-Austin is an EEO/AA Employer.

GLOBAL BIODIVERSITY INFORMATION FACILITY

Executive Secretary

The Global Biodiversity Information Facility (GBIF) seeks a dynamic and visionary individual for the position of Executive Secretary. With the founding Executive Secretary ending his term on 31 October 2007, the Governing Board of GBIF invites applications from suitably qualified individuals to take GBIF forward in its next phase of development.

GBIF is a growing international organisation with 47 countries and 35 international organisations working on the overall mission of making the world's primary biodiversity data freely available through the internet to advance science and increase the use of data for societal benefits at the national, regional and global scale. The aim is to develop an infrastructure facility for access to biodiversity data that is fully interoperable with other environmental and scientific data whereby users all over the world will be able to make more advanced and complex analyses in support of innovation and decision making on many issues relating to biodiversity. Currently GBIF allows access to more than 800 dataset holding more than 100 million records.

GBIF's progress since its inception in 2001 and its strategy for the next five years and work plans can be seen at **www.gbif.org**

GBIF seeks an Executive Secretary with international standing who can work with all participants, inspire and lead the staff at the GBIF Secretariat and establish the key relationships needed in the new phase. GBIF has successfully completed the prototype phase and is now implementing a fully operational system with the challenging overall goal of making GBIF an indispensable tool for decision-makers, scientists, information users and data providers working on biodiversity issues. The Executive Secretary will be responsible to the Governing Board for implementing the GBIF plans and developing new strategic relationships to expand GBIF's influence and directions.

The Executive Secretary will head the Secretariat based in Copenhagen, Denmark and be offered an initial contract of 5 years. A competitive salary will be negotiated with the successful candidate who will benefit from the diplomatic status of the GBIF Secretariat.

The Executive Secretary is expected to take up office on 1 November 2007 or shortly thereafter. Suitable candidates will have wide experience in international activities relevant to the goals of GBIF and have demonstrated leadership in a collaborative and networked environment.

Any enquiries should, in the first instance, be directed to Professor David Penman, Chair of the GBIF Governing Board, at **david.penman@canterbury.ac.nz**

More information on the position and instruction for application is available on the GBIF website: **http://www.gbif.org/executive_secretary**

Applications close on **28 February 2007**.

Biology Institute and Assistant and Full Professors, Systems Biology
University of California, Merced

UC Merced is the 10th UC campus and the first new US research university of the 21st century. UC Merced is located at the base of the Sierra Nevada foothills, near Yosemite and the San Francisco Bay Area. The Schools of Natural Sciences and Engineering at UC Merced seek applicants for faculty positions in Systems Biology at the level of assistant professor (tenure track) and full professor (tenured), who will serve as the Founding Director of the new Biomedical Sciences and Systems Biology Institute. The Director will be an established investigator in systems biology, with a strong track record in extramural support, mentoring, and inter-disciplinary and collaborative research, and demonstrated management, strategic planning and fundraising skills. The Director will hold the Bizzini Family Endowed Chair.

Systems biology is a research approach that uses comprehensive datasets and multiple types of analysis to relate the overall function of an organism, organelle, or regulatory pathway to the underlying biochemical or biophysical processes, with an ultimate goal of a predictive understanding of the system's behavior. Applications of special relevance to research emphases at UC Merced include the mechanisms of cell fate decisions, complex diseases such as diabetes or inflammatory disorders, and microbial systems relevant to human disease.

Candidates for the assistant professor and professorial ranks must have a Ph.D. or equivalent, a record of research, publication, and teaching commensurate with a faculty appointment at UC, and a strong interest in creating a curriculum characterized by strong cross-disciplinary links. Applicants are sought in the areas of experimental or computational systems biology. For more information or to apply see:
http://jobs.ucmerced.edu/n/academic/position.jsf?positionId=732
http://jobs.ucmerced.edu/n/academic/position.jsf?positionId=733

For further information contact **David Ojcius, School of Natural Sciences at dojcius@ucmerced.edu.**

UC Merced is an Affirmative Action Equal Opportunity Employer

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WESTERN CONNECTICUT STATE UNIVERSITY

School of Arts & Sciences
Department of Biological and Environmental Sciences
Chairperson
Academic Year 2007-08

New academic programs, new construction projects, including a new "state of the art" student center, and increasing student enrollment all serve to make Western Connecticut State University a vibrant, active campus serving 6,000 undergraduate and graduate students. WestConn is one of the four comprehensive universities that comprise the Connecticut State University System. The main campus is located in Danbury, 50 miles north of New York City. The University is divided into the School of Arts & Sciences, the School of Professional Studies, the Ancell School of Business and the newly created, School of Visual and Performing Arts. Additional information about WestConn is available at **www.wcsu.edu**

The Department of Biological and Environmental Sciences seeks candidates for a tenure track department chairperson at the level of Associate Professor. The Department is housed in a new Science building with extensive modern facilities and equipment. There are 10 tenure-track faculty members serving 160 undergraduate biology majors, 30 MA students in a part-time graduate program, and a large number of nursing students in several of their required courses. We are seeking a chairperson to provide dynamic leadership and program development in biotechnology and related areas. The successful candidate will foster faculty development and scholarship, recruitment of students, and interaction with the local community, as well as manage routine administrative duties and effectively represent the department to the university administration. Some of the candidate's time will be devoted to teaching and research involving undergraduate or M.A. students.

Qualifications: Candidates must have a Ph.D. or equivalent terminal degree, administrative experience at the department level, documented teaching excellence, and a specialization related to biotechnology.

Salary and Benefits: WestConn offers competitive salaries commensurate with candidates' experience and a comprehensive benefit package. Additional information can be found on our website at **http://www.wcsu.edu/working**

Application Process: Interested candidates should submit a cover letter, a current vitae, 3 letters of recommendation, and statements of administrative philosophy, teaching philosophy, and research interests. Applications should be sent to Dr. Howard Russock, Department of Biological and Environmental Sciences, WCSU, 181 White Street, Danbury, CT 06810. Review of applications begins **December 15, 2006** and continues until the position is filled.

Western is an Affirmative Action/ Equal Opportunity Educator/Employer.

Positions @ NIH
THE NATIONAL INSTITUTES OF HEALTH

Department of Health and Human Services
National Institutes of Health
Clinical Center

Tenure-track Physician
Clinical Center/Nuclear Medicine Department

This position is located in The Warren G. Magnuson Clinical Center, Nuclear Medicine Department (NMD).

We are seeking a research-oriented physician for a possible tenure-track position. An M.D. or M.D. /PhD with U.S. Nuclear Medicine Board certification and CT training is needed to provide diagnostic and therapeutic nuclear medicine procedures as well as to participate in clinical research protocols of the NIH Intramural Program. U.S. citizenship or permanent residency status is required.

Please submit your curriculum vitae, bibliography, and a letter describing your clinical, research, and management experience to: **Mrs. Veronica Olaaje, HR Specialist, DHHS, NIH, OD/CSD-E, 2115 E. Jefferson Street, Rm. 2B209 MSC-8503, Bethesda, MD 20892-8503. Phone: 301-435-4748. Email: volaaje@mail.nih.gov**.

Salary is commensurate with experience. This appointment offers a full benefits package (including retirement, health, life and long term care insurance, Thrift Savings Plan participation, etc.). Application packages should be submitted as early as possible, but no later than **December 31, 2006**.

Selection for this position will be based solely on merit, without discrimination for non-merit reasons such as race, color, religion, sex, national origin, politics, marital status, sexual orientation, physical or mental handicap, age or membership or non-membership in an employee organization.

Postdoctoral Fellow Position Available
National Institute of Dental and Craniofacial Research (NIDCR)

The Adult Stem Cell Unit of the Craniofacial and Skeletal Diseases Branch, National Institute of Dental and Craniofacial Research at the National Institutes of Health in Bethesda, Maryland has a postdoctoral fellow position available starting January or February in 2007. The group is interested in the function of adult stem cells in tissue regeneration and immunomodulation. We have a special interest in auto-immune diseases including Sjogren's syndrome and multiple sclerosis. We have been studying the contribution of circulating bone marrow cells to tissue regeneration in health and disease using genetically marked transplanted bone marrow to track cell fate in the central nervous system and in craniofacial structures (salivary gland; oral mucosa; tongue; skin). Candidates with publications demonstrating laboratory experience in molecular biology, histology and tissue culture techniques are preferred. The work involves animal (mouse and rat) experiments. Salary will be commensurate with experience. To apply, please send a cover letter, curriculum vitae, bibliography, references and a brief description of your special interest to:

Éva Mezey, MD, PhDIH, NIDCR, Bldg 49, Rm 5A-76, 49 Convent Drive, Bethesda, Md, 20892, mezeye@mail.nih.gov.

Department of Health and Human Services
National Institutes of Health
Tenure-Track Position

The Division of Intramural Research, National Institute on Deafness and Other Communication Disorders (NIDCD), located in Bethesda, MD, is seeking a tenure-track scientist to establish an independent research program to study molecular and/or cellular mechanisms of hearing and balance. We welcome applications from candidates with a wide range of expertise. Preference will be given to candidates whose experimental approaches complement those of our existing strong programs in the genetics, development and cell biology of hearing. The successful candidate will join a dynamic group of scientists in a growing intramural program that is at the forefront of research on communication disorders.

The NIDCD offers an exceptional working environment including well-equipped research laboratories and numerous opportunities for collaboration. Candidates for this position must possess a Ph.D. and/or M.D., post-doctoral experience, and an outstanding publication record. Salary is commensurate with education and experience.

Please submit a curriculum vitae including bibliography, three reprints of recent relevant publications, statement of research interests, an outline of your proposed research, and the names and addresses of three references to: **Ms. Trudy Joiner, Office of the Scientific Director, NIDCD, 5 Research Court, Room 2B28, Rockville, MD 20850 (joinert@nidcd.nih.gov)**. Applications will be accepted until **December 15, 2006**.

Department of Health and Human Services
National Institutes of Health
National Cancer Institutes

Tenure-Track Principal Investigator, Laboratory of Cellular Oncology and Center for Cancer Research, NCI.

The Laboratory of Cellular Oncology (LCO), Center for Cancer Research, of the National Institutes of Health, invites applications for a tenure track or tenure eligible principal investigator position in the area of research on papillomavirus biology, including vaccines. The LCO, which occupies recently renovated laboratory space, fosters an interactive research environment and the use of diverse experimental approaches and model systems. Applicants should have a Ph.D. and/or M.D. degree, a strong publication record, and demonstrated potential for imaginative research. Salary will be commensurate with education and experience. A one- or two-page statement of research interests and goals should be submitted in addition to three letters of recommendation and a curriculum vitae to: **Theresa Jones, Laboratory of Cellular Oncology, National Cancer Institute, NIH, Building 37, Room 4106, Convent Drive MSC 4263, Bethesda, MD 20892-4263; phone: 301-496-9513; fax: 301-480-5322; email: jonest@DC37A.nci.nih.gov**. Candidates must be U.S. citizens or permanent residents.

Applications must be received by 1**1/22/06**. The National Cancer Institute is an Equal Opportunity Employer. Selection for this position will be based solely on merit, with no discrimination for non-merit reasons such as race, color, religion, gender, national origin, politics, marital status, physical or mental disability, age, sexual orientations, or membership or non-membership in an employee organization.

WWW.NIH.GOV

Department of Health and Human Services
National Institutes of Health
Director, National Center for Research Resources and
Associate Director for Clinical Research (Extramural)

The Office of the Director, National Institutes of Health (NIH) in Bethesda, Maryland, is seeking applications from exceptional candidates for the position of Director, National Center for Research Resources (NCRR). The Director, NCRR, will also serve as the NIH Associate Director for Clinical Research (Extramural). NCRR, with a staff of approximately 100 employees and a $1 billion budget, is the focal point at NIH for biomedical, clinical and translational research resources. The incumbent serves as a principal advisor to the Director, NIH; participates in discussions relative to the development of major policy decisions affecting biomedical, clinical and translational research resources; provides advice and consultation to NIH components, advisory councils and grantee organizations and institutions; and assures that effective administrative procedures are established so that program operations and obligations of government funds and other resources are rendered consistent with statutory and regulatory requirements and within limitations imposed by the Department of Health and Human Services (DHHS) and Executive Branch policies. As Associate Director for Clinical Research (Extramural), the incumbent is expected to provide leadership for clinical research activities across the NIH. This leadership will involve the coordination of clinical research activities to enhance the integration of basic and clinical research. The Associate Director for Clinical Research will work closely with the other Institute and Center Directors to enhance the efficiency and effectiveness of clinical research supported by the NIH. Applicants must possess a Ph.D., M.D., or a comparable doctorate degree in the health sciences field plus senior level scientific experience and knowledge of biomedical, clinical and/or translational research programs in one or more health science areas. Salary is commensurate with experience and a full package of benefits (including retirement, health, life, long term care insurance, Thrift Savings Plan participation, etc.) is available. A detailed vacancy announcement, along with mandatory qualifications and application procedures, can be obtained via the NIH Home Page at: **http://www.jobs.nih.gov** under the Senior Job Openings section. Dr. Stephen Katz, Director, National Institute of Arthritis and Musculoskeletal and Skin Diseases, and Dr. David Schwartz, Director, National Institute of Environmental Health Sciences, will be serving as co-chairs of the search committee. Questions on application procedures may be addressed to **Ms. Regina Reiter at ReiterR@od.nih.gov or discussed with Ms. Reiter by calling 301-402-1130**. Applications **must** be received by **November 27, 2006**.

NIDDK

HEALTH SCIENCE POLICY ANALYST

The National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) is seeking applications from individuals who are currently in post-doctoral positions in biomedical research laboratories, but who wish to make a career change from a laboratory setting. Particularly encouraged to apply are individuals with post-doctoral experience in molecular biology, coupled with demonstrated writing and other communication skills. Incumbent will develop a wide range of documents that analyze and present the scientific accomplishments and plans of the NIDDK to public policy makers, voluntary health organizations, and other lay audiences. Incumbent must thus be able to convey in understandable, scientifically accurate, and meaningful terms the contributions of biomedical research to human health. Total salary is competitive and will be commensurate with the experience of the selectee.

Position requirements and detailed application procedures are provided on Vacancy Announcement Numbers: **NIDDK-07-155678-MP and NIDDK-07-155678-DE**, which can be obtained by accessing **WWW.USAJOBS.GOV**. All applications must be received by **12/14/06**. For additional information contact **Karen Page at (301) 496-4232**.

NIDDK

Staff Scientist Position
Chemical Biology Core Facility

With nation-wide responsibility for improving the health and well-being of all Americans, the Department of Health and Human Services (DHHS) oversees the biomedical research programs of the National Institutes of Health (NIH) and those of NIH's research Institutes.

The National Institute of Diabetes and Digestive and Kidney Diseases, a major research component of the NIH and the DHHS, is recruiting for a Staff Scientist within the Chemical Biology Core Facility to join a group of investigators whose research focus is directed toward the discovery of new molecules having therapeutic potential. Collaboration with biologists on new strategies to advance basic research will be an important focus of this effort.

The successful individual will be a PhD or MD scientist with at least five years postdoctoral research experience. Research experience should be in synthetic organic or medicinal chemistry. The applicant must have a publication record demonstrating ability to design and carry out multi-step organic reactions.

Applicants should submit a CV, a brief statement of research experience, and the names of three references to: **Kenneth Jacobson, PhD, Bldg. 8A, Rm. B1A19, NIH, Bethesda, MD 20892-0810; or use e-mail: kajacobs@helix.nih.gov mailto:kajacobs@helix.nih.gov ; or FAX: 301-480-8422**. Applications must be received by **December 15, 2006**.

STONY BROOK

Center for Computational Science
Six Positions • 100 Teraflops

Stony Brook University seeks candidates with outstanding potential for six tenure-track or tenure positions in the area of large-scale computational science. The University is in the process of acquiring a large (100 Teraflops class) supercomputer to serve as the core hardware for a newly formed New York Center for Computational Science (NYCCS). NYCCS will provide a home for supercomputing at Stony Brook University and Brookhaven National Laboratory.

Promising scientists who can demonstrate expertise and interest in high-performance computing and/or its utilization are urged to apply. The University will conduct a broad search ranging from traditional computational areas such as those in the physical and life sciences to emerging areas such as those in the social sciences. A successful candidate will hold a faculty position in a University department relevant to his/her interests and will be affiliated with the newly formed New York State Center for Computational Science. For more information, visit the New York State Center for Computational Science Web site at *www.stonybrook.edu/nyccs*

Required: Ph.D., outstanding research and teaching potential, plus experience in large-scale computational science.

The review of materials will begin December 15, 2006, and continue until the six positions are filled.

To apply, please send a résumé; a statement of research and career goals; a statement of teaching goals; the proposed Stony Brook University departmental affiliation; and the name, institutional address, and e-mail address of three references to: NYCCS Search Committee, Posting Number F-3191-06-11, Stony Brook University, SUNY, Stony Brook, NY 11794-1401

For online application visit: *www.stonybrook.edu/cjo*, posting number F-3191-06-11.
Please request that references send letters to the NYCCS Search Committee address above.

Equal Opportunity/Affirmative Action Employer. Women, people of color, individuals with disabilities, and veterans are encouraged to apply.

Assistant Professor of Neuroscience
Washington State University

Tenure-track, full-time, position available in neuroscience at the rank of Assistant Professor to begin on or about July 2007 in the Department of Veterinary and Comparative Anatomy, Pharmacology and Physiology (VCAPP), and the WWAMI Medical Education Program, at Washington State University. Applicants must have a PhD degree or equivalent in neuroscience, cell biology, bioengineering, anatomy, or a related discipline, and have record of research accomplishment.

The successful applicant will be expected to develop and maintain an extramurally funded research program that complements department strengths in systems and cellular neuroscience. These strengths include sleep physiology and function, neural and endocrine control of food intake and body weight, ion channel molecular physiology and biophysics, and substance abuse. The successful applicant will also demonstrate a commitment to biomedical education, and preference will be given to those with previous experience teaching neuroscience, particularly to medical students, and relevant ability working with diverse populations.

Duties will include teaching neuroscience to medical students and participating in neuroscience training of graduate or undergraduate students.

Washington State University has a vibrant neuroscience community and is located in a region having a high quality of life for those who enjoy the outdoors, the arts, and collegiality of neighbors.

Screening of applications will begin **December 15, 2006**. The application must include a cover letter, curriculum vitae, description of teaching experience and philosophy, summary of research interests and goals, and names and contact information (including email addresses) for three references. Send application materials to: **Neuroscience Search Committee, Department of VCAPP, Washington State University, Pullman, WA 99164-6520** or e-mail **bmorton@wsu.edu**.

EEO/AA

TENURE TRACK FACULTY POSITIONS
ENERGY, ENVIRONMENTAL AND CHEMICAL ENGINEERING

The School of Engineering and Applied Sciences (SEAS) at **Washington University in St. Louis** invites applications for five faculty positions at all levels in the newly constituted Department of Energy, Environmental and Chemical Engineering in the following areas:

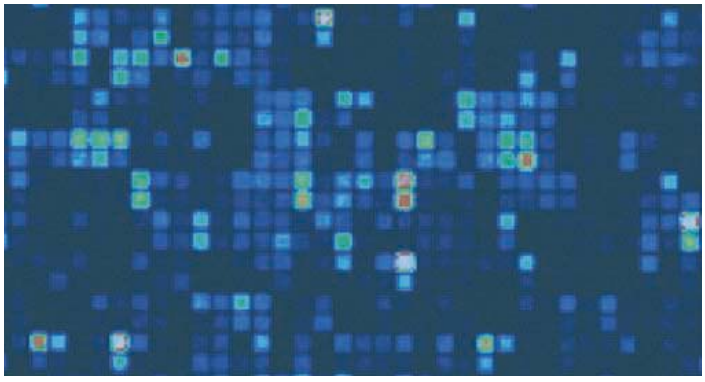
• **Bioenergy and Environmentally Benign Energy Production -** (Up to 3 positions) This is an ambitious program to employ systems-level research approaches to investigate alternate forms of energy production. This program will allow productive interactions between members of SEAS, the College of Arts and Sciences, and the School of Medicine. Preference will be given to individuals with expertise in the following areas: Systems Biology, Bio-Transformation, Metabolic Engineering, Environmentally Benign Energy Production, and Nano-Biotechnology. Applications must be submitted by email as a single file, editable pdf format to **energyfaculty@seas.wustl.edu**.

• **Environmental Engineering Science -** (1 position) This search is open to all areas of aquatic science and technology; however, preference will be given to individuals with expertise in the following areas: Environmental Organic Chemistry, Physical-Chemical Processes in Natural and Engineered Aquatic Systems, Environmental Biotechnology, Environmental Nanotechnology. Applications must be submitted by email as a single file in editable pdf format to **environmentalfaculty@seas.wustl.edu**.

• **Advanced Materials -** (1 position) The School of Engineering and Applied Sciences and the Center for Materials Innovation invite applications for the McKelvey chair in interdisciplinary materials research. Outstanding candidates whose interests fall within a broad range of materials disciplines which include but are not limited to nanoscopic, bio, inorganic AND organic semiconducting, opto-electronic, energy-related, smart, environmental, magnetic and ferroelectric materials. Applications must be submitted by email as a single file in editable pdf format to **materialsfaculty@seas.wustl.edu**.

Review of applications will begin immediately, but applications will be received until the postions are filled. **Washington University in St. Louis, Department of Energy, Environmental and Chemical Engineering, Campus Box 1180, One Brookings Drive, St. Louis, MO 63130 ; 314-935-6070**.

Washington University is an Equal Opportunity and Affirmative Action Employer. Applications from women and under-represented minority groups are strongly encouraged.



Michael, Rituxan® Patient

BECAUSE

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For 30 years, Genentech has been at the forefront of the biotechnology industry, using human genetic information to develop novel medicines for serious and life-threatening diseases. Today, Genentech is among the world's leading biotech companies, with multiple therapies on the market for cancer and other unmet medical needs.

Please take this opportunity to learn about Genentech, where we believe that our employees are our most important asset.

Genentech is dedicated to fostering an environment that is inclusive and encourages diversity of thought, style, skills and perspective. To learn more about these opportunities, please visit **www.gene.com/careers** and reference the Req. #. Please use **"Ad - Science"** when a "source" is requested. Genentech is an equal opportunity employer.

We are currently seeking Senior Scientists and Scientists in our Departments of Immunology, Immunology Diagnostics and Microbial Pathogenesis in our South San Francisco headquarters.

All positions require a PhD, MD or PhD/MD.

Senior Scientist – Immunology, Req. #1000012848
Lead a group of independent Scientists, Research Associates and Postdoctoral Fellows and build a program in tumor immunology and/or autoimmunity. This program will apply human and murine model systems to design and develop therapeutic strategies aimed at regulating the immune system against tumor or self-antigens.

Scientist – Immunology, Req. #1000012849, #1000012850
Develop laboratories aimed at discovering novel immunobiology and translating discoveries into innovative therapeutics. Areas of interest include dendritic cell biology and tumor immunology. Candidates must bring forward novel drug candidates for the development pipeline and publish work in top-tier journals.

Scientist – Immunology Diagnostics, Req. #1000014930
Discover novel immunobiology and translate discoveries into innovative therapeutics and biomarkers. Areas of interest include B-cell immunology and autoimmunity. Candidates should be committed to understanding human disease pathogenesis, mechanisms of therapeutics and identification of markers that may predict clinical efficacy.

Senior Scientist – Microbial Pathogenesis, Req. #1000015208
Scientist – Microbial Pathogenesis, Req. #1000015207
Lead and build research programs in the area of microbial pathogenesis, focusing on understanding the molecular mechanisms by which microbes invade the human host, host-pathogen relationships and drug discovery. Experience in bacteriology and/or virology is required.

Senior Scientists should have an established reputation in leading a research laboratory in academia or industry, proven sustained record of outstanding research performance and major scientific accomplishments. **Scientists** should have postdoctoral experience with an outstanding record of scientific accomplishment in modern human and/or murine immunology as evidenced by the quality of publications.

Genentech was named #1 on Science Magazine's 2006 "Top Employer" list.

Science 2006 TOP EMPLOYER

Genentech
IN BUSINESS FOR LIFE

The **Royal Ontario Museum** (ROM) is Canada's pre-eminent international museum – a superlative showcase of the world's cultural and natural history – a place of discovery, learning, inspiration and imagination, for visitors and staff alike.

Ichthyologist (Freshwater Fishes)

Join us in this entry-level Associate Curator of Ichthyology (equivalent to Assistant Professor) position, to conduct field and collections-related research on freshwater fishes. In addition to developing a program of externally funded scholarly research and publications, you will curate and continue building the disciplinary collection of freshwater fishes, as well as participate in developing and rotating new permanent galleries, travelling exhibitions and other public programming. Your PhD in Systematic Ichthyology is accompanied by expertise in evolutionary biology, historical biogeography, and phylogenetic methods for analysing DNA or morphological data sets. You have a record of scholarly publication in peer-reviewed journals, and are eligible for NSERC funding in support of research, and qualified for cross-appointment to the University of Toronto. Experience in a museum or equivalent environment is preferred.

Salary and rank are commensurate with experience, as stipulated in the Collective Agreement between the ROM and ROM Curatorial Association. Applicants should submit a curriculum vitae, a summary of their research and an outline of their proposed research, and should arrange to have three confidential letters of recommendation sent on their behalf, by **December 29, 2006,** to: **Royal Ontario Museum, Human Resources Department, 100 Queen's Park, Toronto, Ontario, M5S 2C6. Fax: 416-586-5827.** All qualified candidates are encouraged to apply; however, Canadians and permanent residents will be given priority.

Welcome to the new ROM

www.rom.on.ca

ROM Royal Ontario Museum

University of Colorado at Denver and Health Sciences Center Vice Chancellor for Research

The University of Colorado at Denver and Health Sciences Center (UCDHSC) is seeking nominations and applications for the position of Vice Chancellor for Research. UCDHSC is the premier urban research university in Colorado, with exceptional academic programs and over $358 million in combined direct and indirect sponsored programs expenditures. The Vice Chancellor for Research provides direction and advocacy for the research endeavors on both campuses of the university.

For a complete announcement, including the job description, required qualifications and application instructions, please visit our website: **http://www.ucdhsc.edu/admin/hr/jobs/faculty/17513.htm**

The University of Colorado is committed to diversity and equality in education and employment.

The Ohio State University

Assistant Professors in Mathematical Biology/ Evolution and Ecology

Department of Evolution, Ecology, & Organismal Biology (EEOB)
Department of Mathematics

The Department of Evolution, Ecology, and Organismal Biology (EEOB) and the Department of Mathematics invite applications for two tenure-track positions as Assistant Professor in Mathematical Biology. While these positions will be joint hires between the two departments, one position will be tenure-eligible in EEOB and the other in Mathematics. Preference will be given to individuals with interests in areas related to strengths within the Department of EEOB, which include ecology, population biology, population genetics, systematics and evolution, fisheries and aquatic ecology, biodiversity conservation, and behavioral biology. The successful applicants will develop strong, externally funded interdisciplinary research programs, train graduate students, and contribute to undergraduate and graduate teaching. The appointees will be part of a growing faculty in the area of mathematical biology at OSU, with opportunities to participate in the activities of the Mathematical Biosciences Institute, an NSF-funded National Institute located at The Ohio State University. Through research and teaching, the appointees will contribute to bridging biology and mathematics at OSU. Exceptional candidates at the rank of Associate or Full Professor also may be considered. Flexible work options are available.

Applicants should submit their curriculum vitae, statement of research and teaching interests, and contact information for three references online to:

THE OHIO STATE UNIVERSITY

https://www.math.ohio-state.edu/applications/eeob/
or by mail to: **The Ohio State University**
Department of Evolution, Ecology and Organismal Biology
Chair, Mathematical Biology Search
318 W. 12th Avenue
Columbus, OH 43210.

Review of applications begins December 15, 2006 and will continue until suitable candidates are hired.

To build a diverse workforce Ohio State encourages applications from individuals with disabilities, minorities, veterans, and women. EEO/AA employer.

University of Cincinnati

Chairperson of Molecular Genetics, Biochemistry and Microbiology
The University of Cincinnati College of Medicine

The University of Cincinnati College of Medicine invites applications and nominations for the position of Professor and Chair, Department of Molecular Genetics, Biochemistry and Microbiology.

The Department has a history of excellence in research and education. The 24 faculty members hold extramural funding that ranks the Department in the top 10 of public medical school genetics departments. Its faculty is recognized internationally for expertise in cell growth and development, microbiology/immunology, molecular genetics of cardiovascular biology, and structural biology. The Department has played a leading role in developing world-class capabilities in gene-targeting, transgenic mouse models and structural biology for the College of Medicine. The Department also plays a key role in interdisciplinary programs within the College such as cardiovascular sciences, microbiology and obesity/metabolism. The College of Medicine ranks in the top 50 of all research-oriented medical schools, with more than $250 million in sponsored program awards.

We seek candidates with Ph.D. and/or M.D. degrees who are internationally recognized for their research program and have a strong commitment to teaching. The successful applicant will be a versatile scientist who can enhance the activities of the various research groups in the Department. He/she must be a skilled communicator with a clear vision for the future growth and development of a strong multidisciplinary biomedical basic science department that works closely with other research and educational units in the college.

Interested individuals should submit a Curriculum Vitae and the names of three references to: **The Office of Faculty and Administrative Affairs, The University of Cincinnati College of Medicine, 200 Albert Sabin Way, Suite 1200, Holmes, Cincinnati, Ohio 45267-0554, ATTN:MolGen Search;** or e-mail: **marianne.niehaus@uc.edu.** Review of applications will commence immediately and continue until the position is filled.

The University of Cincinnati is an Affirmative Action, Equal Opportunity Employer. Women, minorities, disabled persons, Vietnam era and disabled veterans are encouraged to apply. The U.C. Medical Center is a smoke-free work environment.

USDA ARS Agricultural Research Service
www.ars.usda.gov

The U.S. Department of Agriculture, Agricultural Research Service, San Joaquin Valley Agricultural Research Center, Parlier, California, invites applications for two Research Scientists (GS-12/13/14; $62,291.00-$113,791.00 per annum).

Research Plant Pathologist: Incumbent conducts research independently and as part of a team on the biology of *Xylella fastidiosa* (Xf) strains that cause Pierce's disease (PD) of grapevines, almond leaf scorch, oleander leaf scorch, alfalfa dwarf and other xylellae and exotic and invasive diseases of agronomic, horticultural and ornamental crops (announcement ARS-X7W-0038).

Research Entomologist: Incumbent conducts research independently and as part of a team on the biology and ecology of insect vectors of *Xylella fastidiosa* (Xf) strains that cause Pierce's disease (PD) of grapevines, almond leaf scorch, oleander leaf scorch, alfalfa dwarf and other diseases of crops (announcement ARS-X7W-0039).

Candidate for each position must have the ability to formulate new concepts and hypotheses, and develop new techniques and approaches to solve field problems. Each position is a competitive, permanent appointment and U.S. citizenship is required. Vacancy announcements and where to apply can be found at **www.usajobs.com** or contact **Denice Chambers (559) 596-2960.** Closing date for applications is **January 16, 2007** for both positions.

The USDA is an Equal Opportunity Provider and Employer.

ASSISTANT PROFESSOR VERTEBRATE DEVELOPMENTAL BIOLOGY

The University of Texas at Austin invites applications for a tenure-track position as an Assistant Professor in the Section of Molecular Cell and Developmental Biology. We seek an outstanding investigator who will build an active research program addressing important questions in vertebrate developmental biology. Any qualified applicant will be considered, but we are looking in particular for investigators combining developmental biology with cell biological or biochemical approaches. The successful applicant will be joining the Section at an exciting time, and will join several recent hires in cell biology, developmental biology, and related areas. Very generous start-up funds are available, and the successful candidate will also be eligible for affiliation with the Institute for Cellular and Molecular Biology and/or the Institute for Neuroscience. These Institutes provide state-of-the-art facilities and also support excellent graduate programs.

Applications will be considered starting **December 1, 2006** and continue until the position is filled. Applicants should send their CV, a statement of current and future research interests, representative publications, and three letters of reference to:

Chair, Search Committee
ATTN: Maureen Meko
Section of Molecular Cell and Developmental Biology
University of Texas at Austin
BIO 311, 205 W 24th Street
Austin, Texas 78712-0183

Home Pages: **http://www.biosci.utexas.edu/MCDB/**
and
http://www.icmb.utexas.edu/

The University of Texas, Austin is an Equal Opportunity Employer. Qualified women and minorities are encouraged to apply.

McGill

Two Tenure Track Positions in Cell and Developmental Biology

The Department of Biology at McGill University invites applications for two positions in Cell and Developmental Biology. Candidates using fungal (budding or fission yeast) or genetically well-characterized non-mammalian model systems are particularly encouraged to apply, as are applicants focusing on subcellular structures using advanced imaging and microscopy techniques or single-molecule manipulations.

The successful candidates will be joining the Developmental Biology Research Initiative (DBRI), a dynamic, interactive group of researchers working on a range of subjects in yeast, *C. elegans*, *Drosophila*, *Xenopus*, *Arabidopsis* and other model organisms (**http://www.biology.mcgill.ca/DBRI/dbri_home.htm**). The DBRI is completing a $19.8M infrastructure renovation and renewal project, and is an integral part of the McGill University Life Sciences Research Complex. Ample newly renovated or newly constructed laboratory and office space will be provided to the successful candidates. We anticipate that these positions will be filled at the Assistant Professor (tenure-track) level, but applications from more established candidates may be considered for recruitment at the Associate or Full Professor rank. Competitive start-up and equipment funding packages will be available. The successful candidate is expected to contribute to undergraduate and graduate teaching in the department and to maintain an externally funded research program.

Applicants should possess a Ph. D. degree and significant postdoctoral experience resulting in research publications. Persons wishing to be considered for these positions should forward a curriculum vitae, a statement of research interests, a statement of teaching interests, copies of major publications and arrange to have three letters of reference submitted directly to: **Cell and Developmental Biology Search, c/o Ms. Zabrina Kadkhodayan, Department of Biology, McGill University, 1205 Docteur Penfield Ave., Montreal, Quebec, H3A 1B1, Canada.** The application deadline is **31 December 2006**.

In accordance with Canadian immigration regulations, this advertisement is directed in the first instance to Canadian citizens and landed immigrants, however, all qualified candidates are encouraged to apply.

THE UNIVERSITY OF TEXAS SOUTHWESTERN MEDICAL CENTER

ASSISTANT PROFESSORS: The Department of Physiology invites outstanding scientists with Ph.D., M.D., or equivalent degrees to apply for tenure-track assistant professor positions. Candidates who use innovative optical, mechanical, electrical, molecular biological or computational methods with important applications to physiological systems, ranging from individual genes and proteins to cells and organs are encouraged to apply.

These positions are part of the continuing growth of the Department at one of the country's leading academic medical centers and will be supported by significant laboratory space on our new campus, competitive salaries, and exceptional start-up packages. The UT Southwestern is the scientific home to four Nobel Prize laureates, 17 members of the National Academy of Sciences and 19 members of the Institute of Medicine. More than 2,000 research projects are supported by $300 million grant funding annually at UT Southwestern.

Applicants should submit curriculum vitae, a brief statement of research plans, and arrange to have three letters of reference sent to: **James Stull, Ph.D., c/o Gena McElyea, Department of Physiology, The University of Texas Southwestern Medical Center, 5323 Harry Hines Boulevard, Dallas, TX 75390-9040**.

UT Southwestern strongly encourages applications from women, minorities, and people with physical challenges. An Equal Opportunity Employer.

Celsense

A **Senior Scientist** position is available for a biologist at Celsense, Inc., in Pittsburgh, Pennsylvania. The successful candidate will participate in the development and commercialization of in vivo cellular imaging agents for magnetic resonance imaging (MRI) applications.

Requirements include: a Ph.D. in biological sciences or a related field with at least 3-5 years of bench R&D experience; a background in mammalian tissue culture; in vitro toxicity assays; animal models; molecular biology techniques. A background in immunology would be helpful. Candidates should have strong scientific problem solving skills; ability to communicate results in a clear and concise manner verbally and in writing; record of scientific achievement as documented by peer-reviewed journal publications. Scientific project management experience is preferred. Some travel is required. Celsense offers a competitive compensation and benefits package.

Interested candidates should send a resume/CV to:

Charles F. O'Hanlon
President and CEO, Celsense, Inc.
100 Technology Drive, Suite 400
Pittsburgh, PA 15219 USA
or email:charlie@celsense.com

UNIVERSITY OF CALIFORNIA
UC RIVERSIDE

FACULTY POSITION ANNOUNCEMENTS

ASSISTANT PROFESSOR - CONSERVATION ECOLOGY/BIOLOGY
Department of Biology

Applications are invited for a tenure-track 9-month academic position at the assistant professor rank beginning Fall 2007. A Ph.D. in Conservation Ecology/Biology or related field and at least one year of postdoctoral research experience are required. Applicants are expected to develop a fundamental research program in Conservation Ecology/Biology. Applicants with an emphasis in community to landscape or regional ecology are especially encouraged. The position is open to any area of Conservation Biology, but particular emphasis is placed on topic areas focusing on multiple species interactions examined over community to landscape or regional scales.

Potential collaborators within the Department of Biology and the College of Natural and Agricultural Sciences include over one hundred faculty, ranging from conservation geneticists and population biologists to economists and anthropologists. Opportunities also include collaborations with a variety of research centers in the College including the UCR Center for Conservation Biology. Contributions to teaching at the undergraduate and graduate levels are expected, and there are a variety of departmental and interdepartmental programs that provide opportunities for graduate training. Salary is commensurate with education and experience.

Applicants should submit a CV, a statement of research and teaching interests, a few selected reprints, and letters of recommendation from three referees. We encourage submission of these documents as attachments to emails directed to **conservation ecology@ucr.edu.** Paper copies can be mailed to **Conservation Ecology Search Committee, Department of Biology, University of California, Riverside, CA 92521-0334**. Review of applications will begin on **January 1, 2007** and will continue until the position is filled. Appointment can be as early as **July 1, 2007**.

ASSISTANT PROFESSOR - FUNCTIONAL LANDSCAPE ECOLOGIST
Department of Botany and Plant Sciences

Applications are invited for an assistant level position in Functional Landscape Ecology. This is a tenure-track, academic year (9-month) appointment. The position is designed to integrate physiologically based mechanistic studies with spatial heterogeneity and scale effects to understand functional attributes of spatially heterogeneous landscapes. Candidates may have a background in landscape ecology, ecosystem ecology, physiological ecology, or related fields. The candidate is expected to be well-versed in the technological aspects of the field, such as expertise in remote sensing image processing, GIS and spatial analysis, scaling procedures, or stable isotopes. Research may include topics such as vegetation conversion under global change, species shifts or invasions across landscapes, nutrient movement or eutrophication and subsequent biotic impacts, or others.

The successful candidate will hold a faculty position as well as a joint appointment in the Agricultural Experiment Station. Duties will include developing a cutting edge research program in functional aspects of landscape ecology, supervising graduate students, teaching courses in landscape and ecosystem ecology at the undergraduate or graduate level depending on expertise, and service to the Department and University. Opportunities exist for interaction with programs in Conservation Biology and Environmental Sciences as well as computational sciences in the College. The review of applications will begin **January 15, 2007**, with appointment as early as **July 1, 2007**. Applicants must hold a Ph.D. with a minimum of 1-3 years of Postdoctoral experience. Applications will be accepted until the position is filled.

Applicants should submit the following: (1) a curriculum vitae, (2) a brief statement of research and teaching interests, (3) samples of relevant publications, and (4) have three letters of recommendation sent to: **bpssearch@ucr.edu** or mail to **Chair, Functional Landscape Ecology Search Committee, c/o Department of Botany and Plant Sciences, University of California, Riverside, CA 92521-0124**.

Information about these UC Riverside Departments is available at **http://www.biology.ucr.edu/** and **http://www.plantbiology.ucr.edu/** (see also **http://www.ucr.edu/, http://www.cnas.ucr.edu/**, and **http://www.ccb.ucr.edu/**).

The University of California, Riverside is an Affirmative Action Equal Opportunity Employer committed to excellence through diversity.

Tenure Track Faculty Positions
Viral Pathogenesis-Inflammation

Fox Chase Cancer Center is seeking to recruit new tenure track faculty at the level of Associate Member (equivalent to Assistant Professor) or Member (equivalent to Associate Professor) to join the Viral Pathogenesis program.

We are particularly interested in candidates that will use in vivo models of viral infection to investigate mechanisms of one or several of the following: innate immunity, adaptive immunity, tolerance, inflammation or carcinogenesis.

The successful candidates must have outstanding credentials and will be expected to establish a strong, extramurally funded research program. Fox Chase Cancer Center is an independent research institution that offers a generous start up package and a highly interactive environment.

Application package must be submitted electronically to **Skalka.Recruitment@fccc.edu.** Package should include curriculum vitae, a concise research statement describing research accomplishments and future research plans (4 pages maximum) and contact information for at least three referees. Please reference "Viral Pathogenesis faculty position" in your cover letter.

FOX CHASE CANCER CENTER

Fox Chase Cancer Center is an Affirmative Action/ Equal Opportunity Employer and solicits applications from women and under-represented minorities.

Faculty Position in Liver Research

The Division of Gastroenterology, Hepatology and Nutrition and the Liver Research Group of Cincinnati Children's Hospital Medical Center and Research Foundation invite applications for professorship, at either junior or senior level, for research in liver immunology or liver regeneration.

We seek a candidate with an MD or PhD degree and outstanding prospects for research, who will complement existing strengths of the Liver Research Group in the fields of developmental immunology of the liver, autoimmune liver disease and liver regeneration/repair. A successful applicant must be committed to develop and implement independent, externally funded research programs, mentor students and facilitate interdisciplinary research. The successful applicant will be a member of a highly collaborative group of investigators and will have access to outstanding core resources at the NIH-funded Digestive Health Center and Cincinnati Children's Research Foundation.

Cincinnati Children's Hospital Medical Center and Research Foundation are internationally recognized among the nation's top pediatric care and research institutions. The Research Foundation ranks second nationally in NIH funding to full-service children's hospitals. Cincinnati is a friendly, pleasant, affordable city with a great quality of life, including many musical and theatrical programs, professional sports and nearby recreational opportunities.

To Apply:

Academic rank will be determined by the candidate's credentials and laboratory experience. Applicants should submit a curriculum vitae and statement of research interests to:

Dr. Jorge Bezerra, Director of Research
Division of Gastroenterology,
Hepatology and Nutrition
Cincinnati Children's Hospital Medical Center
3333 Burnet Avenue
Cincinnati, OH 45229-3039
Jorge.Bezerra@cchmc.org

Cincinnati Children's® Hospital Medical Center

Visit our website at www.cincinnatichildrens.org

Cincinnati Children's Hospital Medical Center is an Affirmative Action/Equal Opportunity Institution. Women and minorities are encouraged to apply.

CIC nanoGUNE
nanoscience cooperative research center

The Nanoscience Cooperative Research Center **CIC nanoGUNE Consolider** invites applications for a position as

Research Director

CIC nanoGUNE Consolider, located in San Sebastian, Basque Country (Spain) is a R&D Center created recently with the mission of addressing basic and applied strategic world-class research in nanoscience and nanotechnology, fostering high-standard training and education of researchers in the field, and supporting the growth of a nanotechnology-based Basque industry.

The Research Director of nanoGUNE, working closely with the General Director, will be responsible for the design, coordination, and operation of the Research Laboratories. He/she will also serve as the technical agent in the Center's industrial outreach and would lead one of the Research Laboratories.

Candidates should have an outstanding applied-research record in Physics, Chemistry, Biology, and/or Engineering, with an orientation to nanoscience and nanonotechnology, as well as knowledge of the industrial environment and a leadership in the scientific and technological realms. Preference will be given to candidates with a proven record of attracting competitive research funds and of technological transfer initiatives. Proficiency in spoken and written English is compulsory; knowledge of Spanish is not a requirement. Candidates will be appointed with tenure. An attractive remuneration will be offered.

Applicants should forward their CV to **director@nanogune.eu**

Closing date: 10 December 2006

CONSERVATION BIOLOGIST
CAL POLY, SAN LUIS OBISPO

The Biological Sciences Department within the College of Science and Mathematics at Cal Poly, San Luis Obispo, California is seeking a Terrestrial Vertebrate Conservation Biologist for a full-time, academic year, tenure-track position at the assistant professor rank beginning September 2007. Primary teaching responsibilities will include courses in Conservation Biology, Wildlife Biology, Wildlife Management, and other undergraduate and graduate courses as appropriate to background and training. Ph.D. in related field required at time of hiring. The successful candidate must have a strong commitment to undergraduate teaching, curriculum development, and implementation of a student-centered research program.

Experience in Wildlife Habitat Modeling, Endangered Species Management, Remote Sensing, GIS, and/or Metapopulation Ecology, and postdoctoral or equivalent experience desirable. Salary is commensurate with qualifications and experience.

To apply, visit **WWW.CALPOLYJOBS.ORG**, complete a required online faculty application and submit to Requisition **#101129**; attach your curriculum vitae, statement of teaching philosophy, and statement of professional goals. Also mail a hard copy of the above noted documents and arrange to have official graduate transcripts, and three letters of recommendation sent to: **Dr. Michael Yoshimura, Chair, Biological Sciences Department, California Polytechnic State University, San Luis Obispo, CA 93407-0401**. Review of applications will begin **January 15, 2007**. Applicants are strongly encouraged to have all materials submitted by **January 15, 2007**; applications received after this date may be considered. For questions, contact the **Biological Sciences Department** at **(805) 756-5242**.

Cal Poly is strongly committed to achieving excellence through cultural diversity. The university actively encourages applications and nominations of all qualified individuals. EEO.

Systems Biology
University or Toronto

The new Department of Cell and Systems Biology at the University of Toronto invites applications for a tenure track faculty position to be appointed at the Assistant Professor level in the field of Systems Biology to begin July 1, 2007.

We particularly encourage candidates to apply who have demonstrated excellence in addressing fundamental questions in biology using high-throughput approaches or gene/protein network analyses with bioinformatics, genomic or proteomic tools. Our vision is to advance Systems-wide analyses in Plant, Animal or Microbial Biology which complement existing strengths in the department (**www.csb.utoronto.ca**).

Candidates should have at least two years of research experience beyond their doctoral degree. In addition to pursuing a vigorous, internationally recognized research program, the successful candidate will contribute to undergraduate and graduate teaching in the molecular life sciences. She or he would also be expected to network with researchers across campus to take advantage of the extensive resources in Systems Biology at the University of Toronto and its affiliated institutions. A generous start-up package will be provided. Salary commensurate with qualification and experience.

Applicants should arrange to have at least three letters of recommendation sent directly to the address below. In addition, applicants should forward their curriculum vitae, copies of significant publications, and statements of research and teaching interests to the **Chair, Systems Biology Search Committee, Department of Cell and Systems Biology, University of Toronto, 25 Harbord Street, Toronto, Ontario M5S 3G5, Canada** by **December 31, 2006**. Inquiries should be directed to Ulrich Tepass at **utepass@zoo.utoronto.ca**.

The University of Toronto offers the opportunity to teach, conduct research and live in one of the most diverse cities in the world, and is responsive to the needs of dual career couples. The University of Toronto is strongly committed to diversity within its community and especially welcomes applications from visible minority group members, women, Aboriginal persons, persons with disabilities, members of sexual minority groups, and others who may contribute to the further diversification of ideas. All qualified candidates are encouraged to apply; however, Canadians and permanent residents will be given priority.

Reginald A. Daly Postdoctoral Fellowship
Harvard University
Department of Earth and Planetary Sciences

The Department of Earth and Planetary Sciences at Harvard University invites applicants for the Reginald A. Daly Postdoctoral Research Fellowships.

The Department seeks outstanding candidates in the broad field of Earth and Planetary Sciences. **We encourage applications of candidates pursuing field observations, lab-based science, and theory, and interested in geology, geochemistry, ocean, atmosphere and climate dynamics and chemistry, seismology, geophysics, planetary sciences, and other related fields.** These honorific postdoctoral fellowships are awarded for a one-year period, with an anticipated extension for a second year. Daly fellows carry out independent research, yet are encouraged to interact with one or more research groups in the department. Applicants are welcome to contact members of the department before applying. Applications should include a curriculum vitae, names and affiliation of three referees, a one page statement of the applicant's doctoral research, and a one to two page postdoctoral research proposal. Applications are due **January 15, 2007**. Applicants are responsible for contacting the referees to have their letters arrive directly at the address below by the **January 15, 2007** deadline. Send applications (email preferred) to: **Daly Postdoctoral Search Committee, c/o Rady Rogers (rmrogers@fas.harvard.edu), Department of Earth and Planetary Sciences, Harvard University, 20 Oxford Street, Cambridge, MA 02138.**

The annual salary is $52,000 with additional funds of $15,000 available for research support over a two-year period. Applicants should have a recent Ph.D. or should be 2007 degree candidates. Completion of the Ph.D. is required by the time of the appointment. For more information about the department and Daly postdoctoral program, please visit **http://www.eps.harvard.edu/daly.php**.

We particularly encourage applications from women and minorities. Harvard University is an Affirmative Action/ Equal Opportunity Employer.

JOHNS HOPKINS
MEDICINE
SCHOOL OF MEDICINE

Neuroscience Faculty Positions
The Solomon H. Snyder Department of Neuroscience

Applications are invited for tenure-track faculty positions at both junior and senior levels in the Solomon H. Snyder Department of Neuroscience at the Johns Hopkins University School of Medicine. Applicants should have interests in molecular, cellular, developmental, systems or behavioral neuroscience, a Ph.D. or M.D., and a strong record of research accomplishments. Faculty members are expected to establish, or to have active independent research programs and participate in teaching graduate and medical students.

Deadline for applications is **December 15th, 2006**. Please submit a PDF file containing curriculum vitae and a brief description of current and future research interests and arrange for three letters of reference to be sent on your behalf.

Richard L. Huganir, Ph.D.
Search Committee
Department of Neuroscience
The Johns Hopkins University
School of Medicine
725 North Wolfe Street, PCTB 904
Baltimore, Maryland 21205

JHUNeuroscience@jhmi.edu

An EEO/AA Employer.

NICHOLAS SCHOOL OF THE ENVIRONMENT AND EARTH SCIENCES
DUKE UNIVERSITY

Position in Earth System Analysis in Earth and Ocean Sciences (EOS)

Duke University's Division of Earth and Ocean Sciences in the Nicholas School of the Environment and Earth Sciences (NSEES) anticipates hiring a global hydrologist whose research emphasis is on climate change and water resources. We seek a natural scientist engaged in the interdisciplinary field of global hydrology, with a focus on the global water cycle, biogeochemical or geochemical properties of water resources, and/or human impacts from changes in global water systems. We seek a candidate with the ability to work at regional or global scales, using global earth systems models; advanced remote sensing technologies; and/or terrestrial observations of the amount and quality of surface and ground water. The candidate will be expected to work with Duke faculty to enhance existing scientific programs on climate change, water resources and hydrology. Additionally, the successful candidate may choose to work with researchers at the Nicholas Institute on Environmental Policy Solutions to establish an interface between climate change, changes in water cycling and quality and water policy.

The appointment is open at an assistant professor level. Candidates should possess a portfolio of experience and accomplishments, a strong interest in teaching and mentoring students, and the capacity for playing an active role in the School's water and climate change programs. The Nicholas School includes 50 faculty representing a diversity of disciplines. We offer professional and graduate degrees, and we direct Duke's undergraduate environmental programs.

Letters of interest should include a curriculum vitae and names of three references, and be sent to: **Chair, Earth System Analysis Search Committee, Division of Earth and Ocean Sciences, Nicholas School of the Environment and Earth Sciences, Box 90227, Duke University, Durham, NC 27708**. Applications are due by **January 1, 2006**.

Duke University is an Equal-Opportunity/Affirmative Action Employer. Women and minorities are encouraged to apply.

NIEHS
National Institute of Environmental Health Sciences

Department of Health and Human Services
National Institutes of Health
National Institute of Environmental Health Sciences
Research Triangle Park, North Carolina

Editor-in-Chief

The National Institute of Environmental Health Sciences is commencing a search for the next Editor-in-Chief of *Environmental Health Perspectives (EHP)*. *EHP* is a peer-reviewed monthly science journal, publishing a wide range of topics related to the impact of the environment on health and disease. The journal has an impact factor of 5.34 and ranks first among 132 environmental science journals and among 90 public, environmental, and occupational health journals. The journal is international in scope and is distributed in 190 countries. The Editorial Search Committee seeks to identify an active scientist in a field related to the environmental health sciences and with previous editorial experience. The objective is to identify the next Editor-in-Chief by February 1, 2007. This individual will then begin working with the Interim Editor and *EHP* staff to complete the transition by July 1, 2007.

Letters of interest and plans for *EHP*, along with curriculum vitae, should be submitted by **December 1, 2006** either electronically or by mail to:

William J. Martin II, M.D.
National Institute of Environmental Health Sciences
PO Box 12233, Mail Drop B2-07
Research Triangle Park, NC 27709
E-mail: lloyd3@niehs.nih.gov

DHHS and NIH are Equal Opportunity Employers.

NICHOLAS SCHOOL OF THE ENVIRONMENT AND EARTH SCIENCES
DUKE UNIVERSITY

The Division of Earth and Ocean Sciences in Duke University's Nicholas School of the Environment and Earth Sciences (NSEES) anticipates hiring the second of two Jeffrey and Martha Gendell Chairs in Energy and the Environment. We seek a physical scientist who is a recognized authority on current and future energy resources. This individual's expertise would ideally encompass the availability of energy resources, the technologies and additional resources needed to extract, process, distribute and generate power from them, and the environmental impacts of the resource use. An understanding of the current and future demand for energy resources within the evolving geopolitical landscape of the world is highly desirable. So too are new ideas on the efficient utilization of energy resources. We are equally interested in candidates with a commitment to, and proven record of, interdisciplinary collaboration on problems at the intersection of energy with climate and water.

The appointment is open to all levels: assistant, associate and full professor. Candidates should possess a portfolio of experience and accomplishments commensurate with rank, a strong interest in teaching and mentoring students, and the capacity for playing an active role in the School's Energy and Environment Program. This role will include participating in collaborative initiatives between NSEES and other Duke Schools (Pratt School of Engineering, Fuqua School of Business, the Law School, the Terry Sanford Institute for Public Policy, and Trinity College) which are developing a broad, interdisciplinary program that addresses society's need for affordable, sustainable, safe and clean energy.

The Nicholas School includes 50 faculty representing a divesity of disciplines. We offer professional and graduate degrees, and we direct Duke's undergraduate environmental programs.

Letters of interest should include a curriculum vitae and names of three references, and be sent to: **Chair, Gendell Professorship Search Committee, Earth and Ocean Sciences, Nicholas School of the Environment and Earth Sciences, Box 90227, Duke University, Durham, NC 27708**. Applications are due by **January 1, 2005**.

Duke University is an Equal-Opportunity/Affirmative Action Employer. Women and minorities are encouraged to apply.

南開大學

NANKAI UNIVERSITY

DEANSHIPS & FACULTY POSITIONS

Nankai University was founded in 1919 and is located in Tianjin, a major city and the economic center of northern China. Consistently ranked as one of the top universities in China since it was founded, Nankai University is a key multi-disciplinary university composed of 21 colleges and encompassing both the arts and sciences. The President of Nankai University, Professor Zihe Rao, cordially invites applications from outstanding candidates for the following full-time tenure track positions:

1. Dean & faculty positions in the College of Arts
2. Dean & faculty positions in the College of History
3. Dean & faculty positions in the College of Foreign Languages and Literature
4. Dean & faculty positions in the International College of Chinese Studies
5. Dean & faculty positions in the College of Economics
6. Dean & faculty positions in the Business School
7. Dean & faculty positions in the College of Mathematics
8. Dean & faculty positions in the College of Physical Science
9. Dean & faculty positions in the College of Life Sciences
10. Dean & faculty positions in the College of Information Science and Technology
11. Dean & faculty positions in the Medical College
12. Dean & faculty positions in the Software College
13. Dean & faculty positions in the College of Law
14. Dean & faculty positions in the TEDA College of Applied Physics
15. Faculty positions in the Department of Philosophy
16. Faculty positions in the College of Marxist Education
17. Faculty positions in the College of Chemistry
18. Faculty positions in the College of Environmental Science and Engineering
19. Faculty positions in the TEDA School of Biological Sciences and Biotechnology
20. Faculty positions in the Zhou Enlai School of Government
21. Faculty positions in the Shenzhen Financial Engineering College

Ideal candidates for Deanships will possess excellent credentials in leadership, project management, organization and administration. Ideal candidates for all positions will have knowledge of new and emerging concepts and technologies in their respective disciplines. All candidates will be expected to play a key role in developing research agendas and establishing teaching curriculums in close collaboration with the university. Nankai University places particular importance on providing broad and balanced curriculums with a view to training future leaders.

Candidates should have a doctoral degree (PhD or equivalent) related to their discipline, together with excellent written and interpersonal skills. Candidates should also have significant university teaching experience and a proven track record of academic strength and/or research in their chosen discipline. Candidates will be required to work full-time in Nankai University from commencement of their employment.

How to apply: Interested candidates should send a letter of interest describing their background and experience, a curriculum vitae and three letters of recommendation to: Wang Jun, Zheng Long
Address: Nankai University (Rm. 507, University Office Building), No. 94, Weijin Road, Nankai District, Tianjin 300071, China
Tel: +86-22-23505752, +86-22-81330704;
Fax: +86-22-23508197;
E-mail: zzb@nankai.edu.cn, wanj@nankai.edu.cn

Nankai University is an equal opportunity employer. Positions are open to all qualified candidates, regardless of gender or nationality.

Applications should be received no later than **December 23, 2006**.

www.nankai.edu.cn

UCDAVIS School of Veterinary Medicine

Assistant, Associate or Full Professor of Nutritional Biochemistry (11-mo, tenure track). Salary dependent on qualifications and experience; start-up package. *Required:* PhD and/or DVM or equivalent and advanced training in nutrition, biochemistry or related field; successful research program w/state-of-the-art methods in molecular biology, metabolomics, genomics, and/or proteomics to investigate the interface of intermediary metabolism, nutrition and disease; documented research record/high potential to develop an independent research program related to nutritional biochemistry w/ applications to human and animal diseases; demonstrated teaching experience/aptitude in physiological chemistry, nutritional biochemistry, or nutrition; outstanding ability/potential to acquire extramural research support; excellent interpersonal and communication skills and demonstrated ability to work with others in a collegial, team atmosphere. *Preferred:* Experience in multi-disciplinary approaches and collaborations to study metabolic and chronic disease; metabolic regulation; molecular nutrition; nutritional genetics, or microarray/proteomic studies of effects of diet on gene/protein regulation.

To receive fullest consideration, apply by **January 3, 2007**; position open until filled. Electronically submitted applications encouraged. Submit (1) a letter of intent outlining special interest in the position, overall related qualifications and experience and career goals; (2) cv, and (3) the names and addresses of 3 professional references to:

Dr. Isaac N. Pessah, Chair
c/o Ms. Joan Learned
Dept of Molecular Biosciences
School of Veterinary Medicine
University of California
1 Shields Avenue
Davis, CA 95616-8741
Email: jlearned@ucdavis.edu

UCD is an AA/EOE

MARTIN-LUTHER-UNIVERSITY HALLE-WITTENBERG

The **Institute of Biochemistry/Biotechnology** at the Martin-Luther-University Halle-Wittenberg seeks to fill the position of

Professor (W3) "Plant Biochemistry"

The successful candidate will represent the discipline of Plant Biochemistry within the Institute in both research and teaching. The candidate should have a proven track record of international excellence in a topical research area of plant biochemistry, as evidenced by high ranking publications and external grant funding.
Participation in existing interdisciplinary research programmes within and around Halle is expected, as well as a willingness to establish new collaborative ventures. In particular, the candidate is expected to contribute to and consolidate the existing life sciences research network at the Martin-Luther-University Halle-Wittenberg "Structures and mechanisms of biological information processing" (www.exzellenznetzwerk-biowissenschaften.uni-Halle.de), including the collaborative research centres SFB 648 "Molecular mechanisms of information processing in plants" and SFB 610 "Protein states with relevance in cell biology and medicine". Teaching duties include the training of students in plant biochemistry through both lectures and laboratory courses in a Bachelor/Master framework. Participation in other academic and administrational duties within the Department is also expected.
The Martin-Luther-University Halle-Wittenberg strives to promote equal opportunities in science. Due to the fact that females and handicapped persons are under-represented in science, the Martin-Luther-University Halle-Wittenberg seeks to increase the percentage of female and handicapped scientists and encourages them to apply. If they are appropriately qualified according to the above criteria, they will be given preference by the search committee over other candidates with equivalent relevant qualifications. The salary scale is subject to legal and budgetary constraints of the University.
Applicants should submit a CV, a list of publications, a short exposé of their scientific career, and a concise draft of teaching experiences, ongoing and future research activities and on current grant support. Applications should reach the office of the Dean **no later than six weeks after this posting.**
Martin-Luther-University Halle-Wittenberg, Naturwissenschaftliche Fakultät I (Biowissenschaften), Dekan, 06099 Halle/Saale, Germany

Tulane University Health Sciences Center
TULANE CANCER CENTER

Louisiana Cancer Research Consortium
IN NEW ORLEANS

Endowed Eminent Scholar in Molecular Cancer Pharmacology

Tulane Cancer Center and the Louisiana Cancer Research Consortium (LCRC) seek an outstanding cancer scientist to become an Endowed Eminent Scholar, **The *Joe W. and Dorothy Dorsett Brown Foundation Distinguished Chair in Molecular Cancer Pharmacology***. The eminent scholar holding this tenure track position will be responsible for coordinating basic research leading to the discovery and pre-clinical development of cancer therapeutics. Tulane University Health Sciences Center and Louisiana State University Health Sciences Center in New Orleans have joined together to develop the LCRC, with the goal of achieving NCI designation as a Comprehensive Cancer Center. Continuing funding from a new state tax on cigarettes and significant financial commitment by both Tulane and LSU provide **generous resources for the successful candidate to recruit additional faculty members in both basic and clinical sciences. The goal is to establish a world-class program bringing basic research toward clinical testing. The successful candidate will enjoy modern laboratory space, access to shared core resources, and the opportunity to develop further the LCRC Cores.**

Qualified candidates should forward CV and three letters of reference to: Roy S. Weiner, M.D., Director, Tulane Cancer Center, Tulane University Health Sciences Center, 1430 Tulane Ave., SL-68, New Orleans, LA 70112, rweiner@tulane.edu or Krishna C. Agrawal, Ph.D.,Chairman, Department of Pharmacology, Tulane University Health Sciences Center, 1430 Tulane Ave., SL-83, New Orleans, LA 70112, agrawal@tulane.edu.

The position will remain open until a suitable / qualified applicant has been identified.

An affirmative action / equal opportunity employer.

Bone Marrow Transplant Researcher

The Aflac Cancer Center and Blood Disorders Service at the Emory University Department of Pediatrics and Children's Healthcare of Atlanta is seeking an outstanding laboratory based Investigator at the level of Assistant or Associate Professor (Ph.D. and/or M.D.) to join an expanding research program in the areas of transplant immunology, stem cell biology, graft-versus-host disease or intracellular signaling in the context of blood and marrow transplantation. The successful candidate will join a Division with strengths in stem cell transplantation, oxidant stress, angiogenesis and targeted therapeutics and will be expected to establish an independent research program. The Aflac Cancer Center and Blood Disorders Service is an integral part of the Emory research community and provides a supportive, multidisciplinary environment for developing novel translational therapies for the treatment of malignant and other non-malignant hematologic disorders.

Please direct inquiries with a CV to **KY Chiang, MD, PhD.**
Aflac Cancer Center and Blood Disorders Service,
2015 Uppergate Drive, Rm 464, Atlanta, GA 30322.
Kuang-yueh.chiang@choa.org. Phone (404) 785-1272.

Emory University is an Equal Opportunity, Affirmative Action Employer. Women and members of minority groups are strongly encouraged to apply.

EMORY UNIVERSITY

FACULTY POSITIONS IN SCIENCE AND ENGINEERING
SCHOOL OF EARTH AND SPACE EXPLORATION
ARIZONA STATE UNIVERSITY

ASU ARIZONA STATE UNIVERSITY

In July 2006, Arizona State University launched the School of Earth and Space Exploration (SESE) as part of a university-wide initiative in transdisciplinary research and education. SESE faculty are explicitly organizing their research efforts around "grand challenges" in the earth and space sciences. Success in these endeavors will demand the effective integration of many scientific disciplines, as well as a fusion of science with "collaborative engineering", which we define for our purposes simply as the integration of design and informatics theories from many branches of engineering to facilitate scientific exploration and research on Earth and beyond.

Over the next four years, the School will be hiring a large number of faculty in order to build its capacity for such transdisciplinary research and education. We are now accepting applications from creative researchers and inspired educators as part of the first phase in this process. In all cases, doctoral degrees and an enthusiasm for collaborative research are minimum qualifications for these positions. Opportunities are available for joint appointments with other academic units within the College of Liberal Arts and Sciences and the Fulton School of Engineering. The exact number of hires that will be made during this academic year will depend on administrative discussions currently underway at ASU, and on the quality of the applicant pool. Appointments can be made at any level, from tenure-track Assistant to tenured Full Professor, depending on the experience and qualifications of successful candidates. Cluster hires to build new areas of strength are likely. While we will entertain all applications from candidates in five broad areas – Astronomy and Astrophysics, Atmosphere and Hydrosphere Sciences, Biogeosciences, Engineering for Exploration, and Earth and Planetary Sciences – we note some of our most immediate needs below.

ASTRONOMY AND ASTROPHYSICS: We seek applicants motivated by observational and theoretical studies of the evolution of the universe. Special consideration will be given to applicants who focus their research on the life cycles of stars, the history of solar systems, and the characteristics of extrasolar planets. ASU astronomy and astrophysics faculty currently have access to the Large Binocular Telescope, MMT, Magellan, Bok 2.2m, and other Steward Observatory telescopes, through the Steward Observatory TAC process.

ATMOSPHERE AND HYDROSPHERE SCIENCES: We seek applicants with research interests in the origin and development of the fluid envelopes of Earth and other planets. Special consideration will be given to applicants who focus on the co-evolution of the atmosphere and/or hydrosphere with orogenic systems, on the role of water in planetary surface processes, and on the roles of atmospheric, hydrologic, physical oceanographic, or chemical oceanographic processes on ecosystem evolution.

BIOGEOSCIENCES: We seek applicants who study biological processes in the context of planetary evolution. Specialties of interest include the application of molecular microbial ecology, microbial energy transfer, and molecular biomarkers to studies of active ecosystems and the geologic record.

ENGINEERING FOR EXPLORATION: We seek applicants who enjoy working on challenging engineering problems relating to scientific research and the exploration. Examples include the design of: sensors and sensor networks for monitoring ecosystem processes; novel astronomical or remote sensing instrumentation; scientific experiments for space deployment; robotic devices for exploring of extreme environments on Earth and in space; and autonomous vehicles for scientific research in the oceans, atmosphere, or space. Candidates with degrees in astronautics, electrical engineering, environmental engineering, mechanical engineering, and robotics are especially urged to apply. In addition, we are looking for computer science/informatics faculty with research interests in the design and implementation of creative approaches to the management and visualization of large and diverse scientific datasets.

SOLID EARTH AND PLANETARY SCIENCES: We seek applicants who wish to engage in collaborative, multidisciplinary studies of planets, particularly Earth. We are building faculty teams to focus on two broad research themes. Studies of *planetary interiors* would benefit from new faculty perspectives in mineral physics, high-temperature geochemistry/petrology, geodynamics, seismology, and volcanology. Studies of *crustal and surface processes* would engage new faculty with specialties in cosmogenic radionuclide dating, geodesy, high-temperature geochronology, modern and ancient sedimentary processes, remote sensing, and soil science.

Applications must include: 1) a cover letter that includes a description of how your future research and teaching will contribute to the overall goals of the new School; 2) a current CV; and 3) the names, email addresses, and telephone numbers of three references. **Consideration of applications will commence on 1 December 2006, but applications will continue to be considered bi-monthly until the search is closed.** All Phase I hiring decisions will be completed by Summer 2007.

Inquiries and applications must be addressed to **Kip Hodges, Director of the School of Earth and Space Exploration, and submitted electronically to sesenewfac@asu.edu.** Submissions in pdf format are preferred.

ASU is an equal opportunity/affirmative action employer that actively seeks diversity among applicants and promotes a diverse workforce.

GRADUATE PROGRAM

University of California San Diego

Ph.D. program in PLANT SYSTEMS BIOLOGY at UCSD, SALK Institute and TSRI NSF IGERT GRADUATE TRAINING PROGRAM

This interdisciplinary training program will train graduate students with different backgrounds at the interface of biological systems modeling, computational genomics and plant sciences and will position Ph.D. students at the frontier of systems biology. The program will include focused mentoring of each student in two labs by two advisors from distinct disciplines (e.g. Systems Engineering/ Bioinformatics, Bioenergy and Plant Biology). 30 internationally leading laboratories in diverse disciplines are participating in this new program. For further information see: **http://biology.ucsd.edu/psbigert** .

Highly qualified candidates with diverse backgrounds and degrees in computer sciences, engineering, biology, physics, biophysics, mathematics, chemistry, or related subjects are invited to apply by **December 11, 2006**. On-line Application submission preferably to **http://www.biology.ucsd.edu/grad/index.html** or alternatively applicants can apply to the UCSD Bioengineering or Computer Sciences and Engineering Graduate Programs. Each application should indicate your interest in the Plant Systems Biology Program. For further information contact: Program Directors **Julian Schroeder** and **Steve Briggs** (**jischroe@biomail.ucsd.edu**, **ebobkova@ucsd.edu**).

ASSISTANT/ASSOCIATE TENURE TRACK POSITIONS IN BASIC BIOMEDICAL SCIENCES AT THE UNIVERSITY OF TEXAS MEDICAL SCHOOL AT HOUSTON

The University of Texas Medical School at Houston, one of six schools within the renowned UT Health Science Center at Houston, is initiating a multi-year program to recruit the best basic biomedical scientists, numbering approximately 14 over the first two years with similar numbers over the next several years. A new, state-of-the-art 200,000 square-foot research building will be ready for occupancy in the Fall of 2007 to help initiate this program.

Located in the world famous Texas Medical Center, which includes among others, the Brown Foundation Institute of Molecular Medicine, the University of Texas M.D. Anderson Cancer Center, the Texas A&M Institute of Bioscience and Technology and Baylor College of Medicine, the **UT Medical School** is well positioned to offer to successful candidates appointments in one of four basic science departments (Neurobiology and Anatomy, Microbiology and Molecular Genetics, Integrative Biology and Pharmacology, and Biochemistry and Molecular Biology). Cross appointments and other affiliations are possible, such as in clinical departments or in programs which may include: structural biology, membrane biology, molecular pathogenesis, microbial cell signaling and genomics, human genetics, neurobiology of development, biomedical engineering, as well as many other programs are available to the successful applicant.

These positions afford competitive salaries and attractive start-up packages with unparalleled opportunity to interact with scientists throughout the UT Health Science Center, and the Texas Medical Center in general, as well as Rice University and the University of Houston. Successful applicants should have an M.D. or Ph.D. and postdoctoral experience, and will be expected to develop a nationally competitive research program, a successful graduate student mentoring program, and to engage in the teaching and training of graduate and medical students.

This is an unprecedented opportunity for the next generation of biomedical scientists. Please send current curriculum vitae and a statement of research interests as well as the names of at least three referees to Samuel Kaplan, Ph.D., Search Committee Chair. Applications should be directed to:

Samuel Kaplan, Ph.D.
Microbiology & Molecular Genetics
The University of Texas Medical School
6431 Fannin St., MSB 1.206
Houston, TX 77030-1501
E-mail: carolyn.love@uth.tmc.edu

The University of Texas Health Science Center at Houston is an EO/AA Employer. M/F/D/V. This position is subject to Texas Education Code § 51.215. A background check will be required for the final candidate. Women and minorities are encouraged to apply.

PICTURE YOURSELF AS A AAAS SCIENCE & TECHNOLOGY POLICY FELLOW!

Advance your career and serve society by plugging the power of science into public policy. Year-long Science & Technology Policy Fellowships offer opportunities in six thematic areas: Congressional • Diplomacy • Energy, Environment, Agriculture & Natural Resources • Global Stewardship • Health, Education, & Human Services • National Defense & Global Security.

Work in Dynamic Washington, D.C.
Since 1973, AAAS Fellows have been applying their expertise to federal decision-making processes that affect people in the U.S. and around the world. A broad range of assignments is available in the U.S. Congress and executive branch agencies.

Join a Network of Nearly 2,000 Fellows.
AAAS Fellows benefit from a growing and diverse network of colleagues. Applicants must hold a PhD or equivalent doctoral-level degree in any physical, biological, medical/health, or social science, or any engineering discipline. Individuals with a master's degree in engineering and three years of post-degree professional experience also may apply. Federal employees are not eligible and U.S. citizenship is required.

Apply Now!
The application deadline for the 2007-2008 Fellowships is 20 December 2006. Fellowships are awarded in the spring and begin in September. Stipends range from $67,000 to $87,000, depending on experience.
To apply: fellowships.aaas.org

Enhancing Public Policy, Advancing Science Careers

Krista Donaldson, PhD

Mechanical Engineering, Stanford University.

2004-2005 AAAS Fellow at the U.S. Department of State, Bureau of Near Eastern Affairs, Iraq Desk, Economic Section.

Now a research associate at Stanford University's Center for Design Research.

AAAS
ADVANCING SCIENCE, SERVING SOCIETY

TENURE TRACK FACULTY POSITIONS
Cell Biology and Biophysics

The Division of Cell Biology and Biophysics in the School of Biological Sciences at the University of Missouri-Kansas City is expanding its ongoing faculty search to include all faculty ranks and all areas of the School's research emphases, including microbial genetics, molecular/cell biology and biochemistry/biophysics. Appointments will be considered at the ranks of **ASSISTANT, ASSOCIATE** and **FULL PROFESSOR**, commensurate with applicant experience and accomplishments. Successful candidates will be expected to maintain an extramurally funded research program and contribute to the teaching mission of the School in one or more of the following areas: microbiology, molecular biology, cell biology, biochemistry, biophysics. We seek outstanding scholars with demonstrable achievements in research, teaching experience in an English-language institution, and exemplary communication and supervisory skills. The UMKC School of Biological Sciences offers competitive salaries and start-up funds as well as appropriate laboratory space in the Biological Sciences Building, which houses all School faculty and core facilities **http://sbs.umkc.edu/facilities/index.html**). Applications – including a *curriculum vitae*, reprints of publications, summary of present and future research plans and three letters of recommendation (to be solicited by the applicant) – should be forwarded to: **CBB Search Committee, Division of Cell Biology and Biophysics - BSB 403, University of Missouri-Kansas City, 5100 Rockhill Road, Kansas City, MO 64110-2499**. Review of applications is ongoing and will continue until the positions are filled. *EO/AA Employer.*

GEORGIA CAMPUS
PCOM

Faculty Positions - Assistant Professor
(Associate considered)

Georgia Campus – PCOM is seeking candidates for the following full-time faculty positions for its Division of Basic Sciences. GA-PCOM teaches an integrated medical curriculum to osteopathic medical students and has an evolving graduate program in biomedical sciences; candidates for these positions will be expected to make contributions to the teaching of master's level candidates in our biomedical sciences graduate program. Candidates will be expected to engage in scholarly activity by engaging in research activities that will support graduate program development including mentoring of students, publication of works and the pursuit of extramural funding to support an independent research program. Candidates for each position must have an earned PhD degree in the respective field or closely related area; three (3) years of postdoctoral experience is required.

Anatomy

Candidates must have the ability to teach medical gross anatomy, including instruction in cadaver-based laboratory. Preference will be given to individuals with teaching experience in neuroanatomy, histology, embryology and/or biomechanics/kinesiology.

Physiology

Candidates should be broadly trained with emphasis in GI or neurophysiology. Must have interest in developing viable research program.

Microbiology

Prefer training in bacteriology but will consider other areas. Major teaching areas will be in bacteriology and mycology. Broad experience in other areas a plus.

The Georgia Campus of PCOM is Georgia's newest medical college with a total enrollment of 170 students in its first and second year DO classes and 54 biomedical sciences graduate students. The campus is located in beautiful Suwanee, Georgia, just 38 miles from the airport and 33 miles to downtown Atlanta.

Candidates should send letter of interest and curriculum vitae to:**Philadelphia College of Osteopathic Medicine, Human Resources Department, 4190 City Avenue, Philadelphia, PA 19131, Fax 215-871-6505 or email: hr@pcom.edu** EOE

w w w . p c o m . e d u

NORTHWESTERN UNIVERSITY
FEINBERG SCHOOL OF MEDICINE
Statistical Genetics/Population Genetics Faculty Position

Northwestern University Feinberg School of Medicine and the Center for Genetic Medicine seeks to recruit an outstanding individual with research interests in population genetics or statistical genetics for a full-time, tenure-track, faculty position at the level of **ASSISTANT, ASSOCIATE OR FULL PROFESSOR**. Rank of appointment is dependent upon prior experience and research accomplishment. We are especially interested in investigators interested in the genetics of complex human disease. Investigators who complement existing research strengths in cardiovascular disease, cancer, metabolic disorders, obesity and neurodegenerative disease are particularly encouraged to apply.

Candidates should have a Ph.D. and/or M.D. degree and exceptional research potential. Responsibilities of the position are to develop a dynamic, independently funded research program and to participate in medical and graduate student teaching. High quality laboratory space and excellent start-up support will be provided.

Applications must include curriculum vitae, email address, brief statement of proposed research program and three letters of recommendation. Applications will be reviewed on a rolling basis until the position has been filled. Submissions by email are preferred:

Email: geneticsearch@northwestern.edu
Statistical/Population Genetics Search
c/o Center for Genetic Medicine
303 E. Superior St. Lurie 7-125
Chicago, IL 60611

Northwestern University is an Equal Opportunity/Affirmative Action Educator and Employer and invites applications from all qualified individuals. Applications from women and minorities are especially sought.

Two Tenure-Track Biology Faculty
University of California, Merced

UC Merced is the 10th UC campus and the first new US research university of the 21st century. UC Merced is located at the base of the Sierra Nevada foothills, near Yosemite and the San Francisco Bay Area. The School of Natural Sciences at UC Merced seeks applicants for two faculty positions in the biological sciences at the assistant professor level. Candidates must have a Ph.D. or equivalent, a record of research, publication, and teaching commensurate with a faculty appointment at the UC, and a strong interest in creating a curriculum characterized by strong cross-disciplinary links. Applicants are sought in the following three areas (more detailed descriptions provided at the web links listed):

1. **Bioinformatics:** The ideal candidate will develop state-of-the-art research on integrative data analysis and interpretation using mathematical and statistical models in biological systems. The research emphasis should be on integrative approaches, such as systems biology, networks analysis, comparative genomics, computational biology and bioinformatics that address fundamental biological questions in model and non-model organisms **http://jobs.ucmerced.edu/n/academic/position.jsf?positionId=702**. Contact: **Mónica Medina, mmedina@ucmerced.edu**.
2. **Stem Cell Biology:** Applicants are sought in all areas of stem cell biology, including stem cell signaling and cell fate decisions, self renewal and differentiation, hematology, cancer cell biology, developmental biology, and interactions between stem cells and their microenvironment **http://jobs.ucmerced.edu/n/academic/position.jsf?positionId=721**. Contact: **Jennifer Manilay, jmanilay@ucmerced.edu**.

Interested applicants should submit online: curriculum vitae, statements of research and teaching interests, and the names and addresses of five references. Applications will be considered starting on **December 31, 2006**.

UC Merced is an AA/EOP Employer.

POSITIONS OPEN

ASSISTANT/ASSOCIATE PROFESSOR
Drug Delivery/Drug Targeting
School of Pharmacy

The University of Southern California Department of Pharmacology and Pharmaceutical Sciences **(website: http://www.usc.edu/schools/pharmacy/departments)** invites applications for an Assistant/Associate Professor position, tenure-track or tenured, in drug delivery/drug targeting. Candidates should have a doctoral degree and postdoctoral experience in pharmaceutical sciences, pharmacology, or related disciplines. Candidates with expertise in applied targeting strategies, such as gene and protein drug delivery, liposomal/micelle-based delivery, application of nanotechnology to drug delivery, solid phase systems, or membrane transporters are especially encouraged to apply. The successful candidate is expected to develop a strong research program with extramural funding that complements and expands existing departmental strengths in drug design and discovery, epithelial cell biology, membrane trafficking, drug delivery, pharmacokinetic imaging, neurobiology, mitochondrial function, and aging, and is expected to show a strong commitment to teaching.

The University of Southern California offers cutting-edge opportunities for multidisciplinary, interdisciplinary, and translational research collaborations, including an NCI-designated Comprehensive Cancer Center, an NIH-sponsored Liver and Gastrointestinal Diseases Center, the NIH-sponsored Doheny Eye Institute, the USC Provost's Initiatives in Biomedical Imaging Science, Biomedical Nanoscience, and Neuroscience, graduate programs in Drug Discovery and Regulatory Sciences, and a new Center for Stem Cell and Regenerative Medicine. The University also offers access to one of the widest varieties of affiliated private and public hospitals in the United States **(website: http://www.usc.edu/health/ClinHospPharm.html).**

Candidates should send the names of three references, curriculum vitae, and a summary of research accomplishments and future research and educational goals to: **Curtis Okamoto, Ph.D., Chair, Drug Delivery Search Committee, University of Southern California School of Pharmacy, 1985 Zonal Avenue, Los Angeles, CA 90089-9121,** or **e-mail: cokamoto@usc.edu.** Review of applications will begin immediately, and will continue until the position is filled. *The University of Southern California is an Equal Opportunity/Affirmative Action Employer and encourages applications from women and minorities, and provides reasonable accommodation to individuals with known disabilities.*

SENIOR STAFF ASSOCIATE

A **TECHNICAL POSITION** is available in the Department of Neurology, Columbia University.

Applicant will provide support and participate in electrophysiology research within the Comprehensive Epilepsy Center. The position involves developing visualization and analysis tools for electroencephalogram recordings, applying statistical and signal processing methods to the analysis, and presentation of results. Research related to subpial microelectrode grid project, and seizure localization in intracranial patients using local synchrony measures.

Successful applicant should be a Biomedical Engineer with background in signal processing and development of intelligent algorithms. Should have expertise in wavelets, fuzzy logic, and feature extraction. Knowledge of Matlab, C, and LabView preferred. Applicants must have a Ph.D. in electrical engineering or computer science.

Send letter of inquiry and curriculum vitae to:

Dr. Ronald G. Emerson
The Neurological Institute
710 West 168th Street
New York, NY 10032

Columbia University is an Equal Opportunity Employer.

VICE PRESIDENT GLOBAL MARINE STRATEGIES
Conservation International

To serve as **CHAIRPERSON** of the Global Marine Network that provides strategic direction, focus, and leadership for Conservation International's Global Marine activities and planning.

Education qualifications: Advanced degree in marine sciences required. Required skills and capabilities: Strong technical/scientific background in marine conservation science. Proven ability to lead networks of individual professionals, teams, and organizations in a wide array of disciplines. Proven experience and excellence in nonprofit leadership and management. Ability to create innovative strategies, partnerships, and programs. Excellent skills and ability in organizational management and communication. Ability to collaborate with senior levels of management. Ability to build organizational capacity, lead, and align teams.

Preferred skills and capabilities: Demonstrated fundraising experience at eight-figure level. Ability to work in a multidisciplinary and multinational team to successfully integrate marine conservation. Demonstrable interpersonal skills.

Years of experience: 15 plus years of work in marine conservation and/or policy issues.

Applicants should send cover letter and curriculum vitae, or follow resume application procedure. Conservation International is committed to saving our environment. If you are able, please submit your application electronically! Applicants should apply online by visiting **website: http://www.conservation.org/xp/CIWEB/about/jobs/vp_global_marine_strategies.xml,** or you may send your application to: **Conservation International, Human Resources, 1919 M Street N.W., Suite 600, Washington, DC 20036.** No telephone calls please. *Conservation International is an Equal Opportunity Employer.*

EXECUTIVE VICE PRESIDENT

National Disease Research Interchange (NDRI), a not-for-profit company providing scientists with human biomaterials for research, invites applications for Executive Vice President. The Executive Vice President will possess strong financial and organizational competencies with a minimum of ten years of experience working in a scientific environment requiring business management skills. Requires demonstrated success in negotiating sponsored research agreements, supervising technology joint ventures, and evaluating new science and technology.

Qualified candidates with an advanced degree in medicine or a Ph.D. in the biological sciences or medicine in molecular biology, immunology, genetics, pathology, or a related field are expected to have submitted successful grant applications to NIH and be familiar with NIH reporting requirements and leadership. Superior communication skills required. Computer expertise to include advanced spreadsheet, database, and reporting skills. Must have excellent analytic, writing, and presentation skills. An energetic team player committed to organizational growth and identification of new opportunities is required. Competitive salary and excellent benefits. E-mail curriculum vitae to **e-mail: smcgovern@ndriresource.org,** or fax to **S. McGovern** at **fax: 215-557-7154,** or mail to:

Attn: S. McGovern
1628 John F. Kennedy Boulevard
8th Floor, 8 Penn Center
Philadelphia, PA 19103

CAREER OPPORTUNITY

This unique program offers the candidate with an earned doctorate in the life sciences the opportunity to obtain the Doctor of Optometry (OD) degree in 27 months (beginning in March of each year). Employment opportunities exist in research, education, industry, and private practice. Contact the **Admissions Office, telephone: 800-824-5526** at **The New England College of Optometry, 424 Beacon Street, Boston, MA 02115.** Additional information at **website: http://www.neco.edu, e-mail: admissions@neco.edu.**

ASSISTANT PROFESSOR OF PHYSIOLOGY

Elizabethtown College, located in Central Pennsylvania, currently has an opening in our Biology Department for an Assistant Professor of Physiology. This position requires a Ph.D. Please visit **website: http://www.etown.edu/humanresources** for full ad and application instructions. Application deadline: January 2, 2007. *Affirmative Action/Equal Opportunity Employer.*

ASSISTANT PROFESSOR. The Biochemistry and Cellular and Molecular Biology (BCMB) Department at the University of Tennessee, Knoxville (UTK) seeks to fill a tenure-track faculty position at the Assistant Professor level to begin in August 2007, in the following area: computational modeling and simulation of biomolecular structure, dynamics, and function.

The research associated with the appointments will be performed in the Center for Molecular Biophysics at the UTK/Oak Ridge National Laboratory (ORNL) Joint Institute for Biological Sciences at ORNL. The successful candidate for this position will benefit from interactions with strong research groups within the BCMB Department and in other units on campus and at the Oak Ridge National Laboratory in structural biology, neutron sciences, enzyme mechanisms, proteomics, and computational biology. Particularly strong interactions are expected with research undertaken at the new Spallation Neutron Source and the National Leadership Supercomputing Centre, both at ORNL, and with the ORNL Life, Computational and Physical Sciences Programs. The successful applicant will be expected to develop first-class, externally funded research programs, to provide state-of-the-art training for graduate students and postdoctoral researchers, and to contribute to the teaching mission of the BCMB Department at both the undergraduate and graduate levels. Required qualifications include a Ph.D. and postdoctoral experience in relevant areas of computational molecular biophysics, evidence of significant scientific productivity, and a commitment to an integrated program of teaching and research. The University welcomes and honors people of all races, creeds, cultures, and sexual orientations, and values intellectual curiosity, pursuit of knowledge, and academic freedom and integrity. Interested candidates should send a cover letter, a resume, a description of research experience and of the proposed research program, and the names of three individuals who can provide letters of reference to: **Jeremy Smith, Chair, Faculty Search Committee, Biochemistry and Cellular and Molecular Biology Department, M407 WLS, University of Tennessee, Knoxville, TN 37996-0840.** Review of applications will begin on November 17, 2006, and continue until the position is filled.

The University of Tennessee is an Equal Employment Opportunity/Affirmative Action/Title VI/Title IX/Section 504/ADA/ADEA Institution in the provision of its education and employment programs and services.

VISITING SCIENTIST

(26UC4545) University of Cincinnati College of Medicine, Molecular Genetics.

Perform research into the novel poly (ADP-ribose) polymerases tankyrase 1 and 2.

Minimum qualifications: Ph.D. or M.D. with a strong background in biochemistry and molecular biology and must have experience in DNA cloning, PCR, and tissue culture. Good oral and written English language skills are essential.

For additional information on position 26UC4545 see **website: http://www.jobsatuc.com.** *Equal Opportunity Employer.*

OTOLARYNGOLOGIST

The Section of Otolaryngology - Head and Neck Surgery at Dartmouth-Hitchcock Medical Center seeks a board certified or board eligible Otolaryngologist for a full-time faculty position.The candidate should possess an interest in an academic career and in the education of medical students and residents.This position will combine a general otolaryngology with a subspecialty practice in otology or pediatric otolaryngology. Fellowship training in otology/ neurotology or pediatric otolaryngology is desirable. Research interests will be encouraged. Academic rank will be commensurate with qualifications and experience.

Interested applicants are encourage to send letters of inquiry and CV to:

Daniel Morrison, MD, Chairman
Section of Otolaryngology - Head & Neck Surgery
Dartmouth-Hitchcock Medical Center
One Medical Center Drive
Lebanon, NH 03756
Telephone: 603-650-8123

DARTMOUTH–HITCHCOCK
MEDICAL CENTER

Dartmouth-Hitchcock Medical Center is an affirmative action/equal opportunity employer and is especially interested in identifying female and minority candidates.

www.DHMC.org

Department of Health and Human Services
National Institutes of Health
National Institute on Aging
Intramural Research Program

TENURE TRACK - TRANSLATIONAL INVESTIGATOR

The Laboratory of Clinical Investigation (LCI) of the National Institute on Aging (NIA) is recruiting a translational scientist for a tenure-track position within its Intramural Research Program (IRP). This position is 100% research, includes an attractive set-up package and operating budget, and provides the unique and extensive resources of the NIH. Principal Investigators in the LCI include Drs. Darrell R. Abernethy (Lab Chief), Josephine M. Egan, Richard G.S. Spencer, and Irving W. Wainer.

The successful individual will possess an M.D., M.D.,/Ph.D., or a Ph.D. degree with training and experience in translational research in Immunology/Oncology. The successful candidate will have an established record of scientific accomplishment within the fields of clinical immunology/oncology and a strong publication record.

Salary is commensurate with research experience and accomplishments, and a full Civil service package of benefits (including retirement, health, life and long term care insurance, Thrift Savings Plan participation, etc.) is available.

Additional information regarding the NIA/IRP and the LCI are available at the following Websites:

http://www.grc.nia.nih.gov
http://www.grc.nia.nih.gov/branches/INs./index.html

To apply: Please send a cover letter, curriculum vitae, bibliography, and statement of research interest to: Peggy Grothe, Intramural Program Specialist; Office of the Scientific Director (Box 09); Vacancy # **NIA-IRP-06-10**; National Institute on Aging, 5600 Nathan Shock Drive, Baltimore, MD 21224-6825. Applications must be postmarked by **January 16, 2007.** If additional information is needed, please call 410-558-8012 or email **grothep@grc.nia.nih.gov**

DHHS and NIH are Equal Opportunity Employers

THE STATE UNIVERSITY OF NEW JERSEY
RUTGERS

Faculty Position in Human Genetics
Rank Open

The Department of Genetics (**http://lifesci.rutgers.edu/~genetics**) and the Human Genetics Institute of Rutgers University seek an outstanding scientist to fill one of several new positions in human genetics. Researchers seeking a well-funded, diverse, and interactive department are encouraged to apply. Research areas of interest include but are not limited to: population genetics, computational genetics, developmental genetics, chromatin remodeling and epigenetics, complex disease gene discovery, cancer genetics, neurogenetics and neuropsychiatric genetics, and functional genomics.

Candidates must have either a Ph.D. or M.D., or both, a demonstrated ability to conduct and publish significant independent research, and an interest in teaching at the undergraduate and graduate levels. Senior level candidates must have a strong record of grant support. Appointments will be made at a tenured or tenure-track level, consistent with the candidate's credentials.

Laboratory space will be provided in the newly constructed, state-of-the-art Genetics/Human Genetics Institute on Rutgers' Busch Campus. We are part of a vibrant life sciences community including the Waksman Institute, the Center for Advanced Biotechnology and Medicine, the Center of Alcohol Studies, the Environmental and Occupational Health Sciences Institute, and the Robert Wood Johnson Medical School. The campus is located in central New Jersey, close to New York City, Philadelphia, beaches, and countryside.

Applicants should send a CV, a statement of research interests, and full contact information for three individuals willing to provide a detailed evaluation of the candidate to: **genetics_search@biology.rutgers.edu**, or to **Jay Tischfield, Chair, Department of Genetics, Rutgers University, 145 Bevier Road, Room 136, Piscataway, NJ 08854-8082**. Review of applications will begin **December 1, 2006**. The starting date is flexible.

Rutgers University is an Equal Opportunity/ Affirmative Action Employer.

GRADUATE PROGRAM

FALL 2007 Ph.D. PROGRAMS

THE SCRIPPS RESEARCH INSTITUTE

DOCTORAL PROGRAMS IN THE CHEMICAL AND BIOLOGICAL SCIENCES

The Kellogg School of Science and Technology at Scripps Research Institute will admit highly qualified chemistry and biology students to the La Jolla, California or the Jupiter, Florida campus to study biology, biophysics, chemical biology or chemistry, employing a highly interdisciplinary approach including a customized curriculum. The application deadline for Fall 2007 is January 1, 2007.

Established in 1961, Scripps Research Institute has gained international recognition for basic research in chemistry, structural, molecular and cell biology. Graduate studies at Scripps Research Institute provide an exceptional training opportunity in a uniquely multidisciplinary environment with emphasis on individualized training.

Candidates must have a bachelors degree and a strong background in biology, biophysics, chemistry, or a related discipline. Qualified applicants will be invited to visit the campus of admission. Financial support will be provided to all students accepted into the program.

Individuals interested in applying should visit Scripps Research Institute web sites: www.scripps.edu or www.scripps.edu/florida or contact:

Kellogg School of Science and Technology
The Scripps Research Institute
10550 N. Torrey Pines Rd., (TPC 19)
La Jolla, CA 92037

Tel: 858-784-8469 email: gradprgm@scripps.edu

Accredited by the Western Association of Schools and Colleges, 985 Atlantic Avenue, Alameda, CA 94501 (510-748-9001). The Kellogg School of Science and Technology admits students of any race, color, and national or ethnic origin.

KELLOGG SCHOOL
of science and technology

POSITIONS OPEN

FACULTY POSITION
The School of Civil and Environmental Engineering
Position Title: Environmental Biotechnology

The School of Civil and Environmental Engineering invites applications for a tenure-track faculty position with starting date in 2007. Preferred areas of expertise include microbial ecology as it relates to environmental biotechnology and systems biology. The successful candidate must engage in interdisciplinary activities and apply cutting-edge technologies and approaches that complement existing strength in environmental engineering and environmental microbiology. The position requires development of an extramurally funded, internationally recognized, independent research program and excellent teaching at both the undergraduate and graduate levels. Candidates must have a Ph.D. degree and special consideration will be given to candidates with postdoctoral experience. The rank of **ASSISTANT or ASSOCIATE PROFESSOR** will be considered, depending on the qualifications of the candidate. Screening of applicants will begin immediately and will continue until the position is filled.

Applications: Please apply online at **website: http://www.ce.gatech.edu/jobs/#env.** Applications should include a detailed letter discussing interest, background and experience, curriculum vitae, a summary of research interest, a statement of teaching interests and objectives, and the names and contact information of at least four references.

The Georgia Institute of Technology is an Equal Opportunity/Affirmative Action Employer, and applications from women and underrepresented minorities are encouraged.

ASSISTANT/ASSOCIATE PROFESSOR
University of Illinois
College of Medicine at Rockford

The Department of Biomedical Sciences at the University of Illinois, College of Medicine at Rockford invites applications for a tenure-track position at the rank of Assistant or Associate Professor beginning summer of 2007. We are seeking applicants with expertise in pharmacology, microbiology, immunology, or molecular biology. Strong applicants from other areas such as tropical disease and allergy will also be considered. Ph.D. with demonstrated excellence in research and transferable extramural funds are preferred. Successful candidates will be required to participate in team teaching to medical students in pharmacology, microbiology, and/or medical biotechnology. Submit curriculum vitae, a short statement of research interests, and the names and addresses of three references to: **Dr. Ramaswamy, Department of Biomedical Sciences, College of Medicine at Rockford, 1601 Parkview Avenue, Rockford, IL 61107-1897.** Review of applications will begin December 1, 2006. *The University of Illinois is an Equal Opportunity/Affirmative Action Employer.*

The Biology Department at the State University of New York (SUNY), New Paltz **(website: http://www.newpaltz.edu/biology)** invites applications for a **TENURE-TRACK POSITION** starting August 2007. Applicants must have a Ph. D. in biology with expertise in molecular biology or a related area. The successful candidate is expected to teach molecular biology and genetics, develop a graduate and general education course in her/his area of specialty, and establish an independent research program that involves students. A letter of application, curriculum vitae, representative publications, separate statements of research interest and teaching philosophy, and three letters of recommendation should be sent to: **Search Committee (Position F06-35), Department of Biology, State University of New York, New Paltz, 1 Hawk Drive, New Paltz, NY 12561-2443.**

Deadline: January 12, 2007. Electronic submissions will not be accepted.

SUNY New Paltz is an Affirmative Action/Equal Opportunity Employer/ADA Employer.

FACULTY POSITION
Theoretical Condensed Matter Physics
University of Illinois, Urbana-Champaign

The Department of Physics invites applications for a full-time tenure-track faculty position at the **ASSISTANT PROFESSOR** level in the area of theoretical condensed matter physics, beginning as early as August 16, 2007, or approximate date. An appointment at a higher level will be considered for an exceptionally well-qualified candidate. A Ph.D., or equivalent, is required, along with the ability to teach effectively at both undergraduate and graduate levels and to conduct a vigorous and significant research program. For full consideration for the coming academic year, completed applications must be received before January 16, 2007. Salary will be competitive and commensurate with qualifications. Applicants will go online to **website: https://my.physics.uiuc.edu/join/** to complete and submit an application form. Curriculum vitae, a publication list, a summary of research interests and accomplishments, and the names and addresses of three references will be submitted at that time. If you do not have internet access, please contact the **Physics Departmental Office, telephone: 217-333-3760,** to make other arrangements for submitting your application. *The University of Illinois is an Affirmative Action/Equal Opportunity Employer.*

FACULTY POSITION
ASSISTANT/ASSOCIATE PROFESSOR
Plant Physiology/Eco-Physiology
Colorado State University

Responsibilities are to teach and to develop an externally funded research program that aims to better understand physiological and/or ecological mechanisms by which plants respond to water deficit stress, especially in managed landscape ecosystems. The appointee is expected to: obtain grants and build a fundamental research program, collaborate with others in problem-solving approaches to water as a landscape issue in the West, and teach and mentor undergraduate and graduate students. Applicants must have a Ph.D. in a plant biology discipline relevant to the responsibilities of the position.

The full position description and additional information can be obtained from: **Dr. Stephen Wallner, Head, Department of Horticulture and Landscape Architecture, Colorado State University, 301 University Avenue, Fort Collins, CO 80523-1173.**

E-mail: stephen.wallner@colostate.edu, or see horticulture and landscape architecture **website: http://hla.colostate.edu.** *Colorado State University is an Equal Opportunity/Affirmative Action Employer.*

2007 AMERICAN SOCIETY FOR MICROBIOLOGY/CENTER FOR COMPLEX INFECTIOUS DISEASES POSTDOCTORAL RESEARCH POSITIONS IN MICROBIOLOGY

Positions are available for Postdoctoral Scientists to conduct novel research with the overall objective of developing practical applications of microbiology, immunology, and epidemiology for diagnosis and prevention of infectious diseases. Fellows will perform research at one of the Centers for Disease Control and Prevention locations: Atlanta, Georgia, Fort Collins, Colorado, San Juan, Puerto Rico, or Anchorage, Alaska.

Applications must be submitted electronically.

Application deadline: January 15, 2007.

Website: http://www.asm.org/Education/index.asp?bid=15497. E-mail: fellowships-careerinformation@asmusa.org.

The Institute of Technology of the University of Minnesota, Twin Cities, invites nominations and applications for **HEAD OF THE DEPARTMENT OF MECHANICAL ENGINEERING.** Complete job description and application instructions can be found at **website: http://www.me.umn.edu/.** *The University of Minnesota is an Equal Opportunity Educator and Employer.*

TENURE-TRACK FACULTY POSITION IN STRUCTURAL BIOLOGY
University of Maryland Biotechnology Institute, Shady Grove
Center for Advanced Research in Biotechnology
Center for Biosystems Research

As part of a major new expansion, the University of Maryland Biotechnology Institute (UMBI) invites applications for a tenure-track faculty position **(ASSISTANT PROFESSOR)** in structural biology (X-ray crystallography or nuclear magnetic resonance spectroscopy [NMR]). The successful candidate will be expected to develop a competitive and externally funded research program using structural biology approaches to address contemporary biological questions.

The Shady Grove Campus of UMBI includes scientists from the Center for Advanced Research in Biotechnology (CARB, **website: http://carb.umbi.umd.edu/),** the Center for Biosystems Research (CBR, **website: http://www1.umbi.umd.edu/~cbr/),** and the National Institute of Standards and Technology (NIST). The campus is located in the heart of a major biotechnology community with easy access to the National Institutes of Health and NIST. The successful candidate will benefit from existing strengths in structural biology, biophysical chemistry, and computational biology at CARB, and from research into complex biological systems and pathobiology at CBR. State-of-the art facilities and support for X-ray crystallography and NMR are available at Shady Grove.

Qualifications: Ph.D. in biochemistry or related field, postdoctoral experience and knowledge skills in structural biology. Applicants will be considered who have research interests in any area of contemporary structural biology, including biomedical, plant, or insect biology. Applicants should submit their curriculum vitae (referencing position 300881), a summary of future research plans, and names of three references (PDF file) electronically to **e-mail: carbsrch@umbi.umd.edu** or by mail to: **Structural Biology Search Committee, University of Maryland Biotechnology Institute, Shady Grove, 9600 Gudelsky Drive, Rockville, MD 20850.**

Review of candidates will begin January 1, 2007, and continue until the position is filled.

UMBI is an Equal Employment Opportunity/ADA/Affirmative Action Employer.

A **FUNDED POSITION** is immediately available for a person with knowledge of mouse developmental biology. People knowledgeable about limb, hair follicle, or mammary gland development are encouraged to apply. The project concerns the development of molecular genetic methods to identify, isolate, and characterize putative stem cells in the developing mouse embryos, focusing initially on the mammary gland. It would be helpful, but not required, for the candidate to be knowledgeable of molecular and embryologic techniques such as immunnohistochemistry, in situ hybridization, laser capture microdissection, microarray analysis, PCR, and some mouse manipulations, such as ES cell propagation and transfection. Above all, they should be intellectually curious, productive, and enjoy working in a committed team effort to make this important project succeed. The project is well under way due to the efforts of two excellent junior researchers, and is at an exciting stage that could benefit from a person with mentoring skills as well as the aforementioned background. Applicants should submit current curriculum vitae, names of three references, and an indication of the experimental direction they would like to pursue to: **Geoffrey M. Wahl, Ph.D., Professor, Salk Institute for Biological Studies, Gene Expression Laboratory, 10010 N. Torrey Pines Road, La Jolla, CA 92037 U.S.A. E-mail: katz@salk.edu., website: http://www.salk.edu.** *Equal Opportunity Employer.*

POSITIONS OPEN

CENTER OF EXCELLENCE DIRECTOR

The Department of Veterans Affairs' Central Texas Veterans Health Care System (CTVHCS) seeks a Director for a new Center of Excellence for Mental Health and Post-traumatic Stress Disorder, located at the Waco campus of CTVHCS. The program is on the cutting edge of implementing psychosocial rehabilitation for the seriously mentally ill, and is a teaching hospital of the Texas A&M University HSC College of Medicine. Candidates must demonstrate a sustained, independent funded research program in post-traumatic stress syndrome or psychosocial rehabilitation, as well as the vision to successfully guide the Center of Excellence to national prominence. CTVHCS is currently implementing major funding for a new research program in post-traumatic and developmental stress disorders, and administrative experience in overseeing translational research will be highly regarded. Funding will be immediately available to hire at least two additional experienced investigators and administrative personnel to support the program, with additional support expected as the Center of Excellence is implemented.

Salary is commensurate with qualifications and experience. *Applicants must be U.S. citizens* with a current unrestricted license to practice medicine in the U.S. or an earned Ph.D. degree. The position offers excellent benefits, low cost of living, and no state income tax.

To apply, contact:

Mary Doerfler, Human Resources Specialist (05)
Central Texas Veterans Health Care System
1901 Veterans Memorial Drive, Temple, TX 76504
Telephone: 254-743-0049, fax: 254-743-0007
E-mail: mary.doerfler@med.va.gov

POSTDOCTORAL/RESEARCH SCIENTIST
HIV/AIDS Research Program
New York University
College of Dentistry

The HIV/AIDS Research Program at New York University (NYU) College of Dentistry has several openings for Postdoctoral/Research Scientists. Candidates should have a strong foundation and doctoral degree in virology, HIV pathogenesis, immunology and/or molecular biology. Our research program includes: (1) studies on the mechanism of action of gp340 (DMBT1) as an inhibitor of HIV infection, (2) development of a novel point-of-care diagnostic system for the detection of multiple bacterial and/or viral pathogens, (3) Investigate HIV-1 evolution through genetic recombination, and (4) studies of the kinetics of CTL killing of HIV-1 infected cells.

NYU offers excellent benefits. Salary and research rank will be commensurate with credentials and experience. Please forward resume and names of three references. For projects one and two send contact information to: **Dr. Daniel Malamud** at **e-mail: dmalamud_nyucd@yahoo.com.** For projects three and four, send contact information to: **Dr. David N. Levy** at **e-mail: dnlevy_nyucd@yahoo.com.**

NYU is an Equal Opportunity/Affirmative Action Employer.

The Office of Science, Department of Energy is seeking a motivated and highly qualified individual to serve as the **ASSOCIATE DIRECTOR,** Office of Biological and Environmental Research. As such, you will provide leadership and direction in establishing vision, strategic plans, goals, and objectives for the research activities supported. You may apply through two different methods, one is for a **SENIOR EXECUTIVE SERVICE** appointment and the second is for an **INTERGOVERNMENTAL PERSONNEL ACT** appointment. The announcement number is SES-SC-HQ-005. The announcement opens on November 6, 2006, and closes on December 21, 2006. Visit **website: http://www.usajobs.opm.gov/** for more information and for instructions concerning application procedures.

POSITIONS OPEN

TENURE-TRACK FACULTY POSITION IN METABOLOMICS
University of Maryland Biotechnology Institute, Shady Grove
Center for Advanced Research in Biotechnology
Center for Biosystems Research

Applications are invited for a tenure-track faculty position at the **ASSISTANT, ASSOCIATE, or PROFESSOR** level. The successful candidate will be expected to develop a rigorous, externally funded research program in the field of metabolomics using advanced analytical methods.

The Shady Grove Campus of the University of Maryland Biotechnology Institute (UMBI) is developing an integrated research program in molecular systems biology, bridging the interests of the Center for Advanced Research in Biotechnology (CARB, **website: http://carb.umbi.umd.edu/),** a partnership with the National Institute of Standards and Technology (NIST) and the Center for Biosystems Research (CBR, **website: http://www1.umbi.umd.edu/~cbr/).** Research areas at the Shady Grove Campus include chemical biology, mass spectrometry, structural biology, bioinformatics, experimental and computational biophysics, systems modeling, plant and insect biology. Several new faculty hires are anticipated over the next two years, and a new 140,000 square-feet research building equipped with state-of-the-art facilities has recently opened.

Qualifications: Ph.D. in biochemistry or related field, postdoctoral experience, and knowledge skills in metabolomics. Areas of interest include but are not limited to: metabolite changes in response to disease or environmental stress; applications in functional genomics; metabolic networks; medicinal plant metabolism; development of metabolomic databases. We are particularly interested in applicants who are seeking a highly collaborative research environment.

Applicants should submit their curriculum vitae (referencing position 300879), a summary of future research plans, and names of three references (PDF file) electronically to **e-mail: carbsrch@umbi.umd.edu** or by mail to: **Metabolomics Search Committee, University of Maryland Biotechnology Institute, Shady Grove, 9600 Gudelsky Drive, Rockville, MD 20850.**

Review of candidates will begin January 1, 2007, and continue until the position is filled.

UMBI is an Equal Employment Opportunity/ADA/Affirmative Action Employer.

FACULTY POSITION
Theoretical Nuclear Physics
University of Illinois, Urbana-Champaign

The Department of Physics invites applicants for a full-time **TENURED OR TENURE-TRACK FACULTY** position in theoretical nuclear physics, beginning as early as August 2007. The Univeristy of Illinois, Urbana-Champaign Department of Physics has active programs in both theoretical and experimental nuclear physics. The successful candidate must have the ability to teach effectively at both the undergraduate and graduate levels and to lead a vigorous and significant research program. A Ph.D. or equivalent is required. Salary is open and will be commensurate with qualifications. Submission is via the web **(website: https://my.physics.uiuc.edu/join/).** Applicants will submit curriculum vitae, a list of publications, a brief description of their research and teaching interests and plans, as well as names of three references who can provide letters of recommendation. If you do not have internet access, please contact the **Physics Departmental Office, telephone: 217-333-3760,** to make other arrangements for submitting your application. For full consideration, application materials must be received by January 15, 2007. Applications will be accepted until the position is filled. *The University of Illinois is an Affirmative Action/Equal Opportunity Employer. Minorities, women, and other designated class members are encouraged to apply.*

POSITIONS OPEN

ASSISTANT PROFESSOR IN PLANT BIOLOGY
University of Virginia, Charlottesville, Virginia

The Department of Biology at the University of Virginia has an opening for an Assistant Professor (tenure track) starting August 25, 2007. Applications are invited from outstanding individuals studying fundamental aspects of plant developmental biology at the molecular, cellular, organismal, or systems level. We are particularly interested in candidates with a research program that incorporates genetics, functional genomics, computational biology, proteomics, or metabolomics. Our Department spans a broad range of interests including developmental biology, morphogenesis, neurobiology, biological timing, and evolutionary biology. The successful candidate is expected to establish a vigorous, independent, and externally funded research program and to integrate into undergraduate and graduate instruction and training. A generous startup package and excellent research facilities are available. Applicants must have a Ph.D. degree, suitable postdoctoral or academic experience, and a strong research publication record.

To apply, send curriculum vitae, a statement of current and future research interests, a statement of teaching experience and goals, and the names of three references to: **Chair, Plant Biology Search Committee, Department of Biology, University of Virginia, P.O. Box 400328, Charlottesville, VA 22904 U.S.A.** Application materials can also be submitted via **e-mail: biosearch@virginia.edu.** Review of applications will begin December 1, 2006. The position will remain open until filled.

The University of Virginia is an Equal Opportunity/Affirmative Action Employer.

VERTEBRATE ENDOCRINOLOGIST
Tenure-Track Position

The Department of Biological Sciences at California State University, Chico, invites applications for a full-time, tenure-track faculty position as an **ASSISTANT PROFESSOR** in vertebrate endocrinology to begin fall 2007. Applicants should have a strong background in vertebrate endocrinology and cell physiology. The successful candidate will be expected to pursue an externally funded research program involving undergraduate and Master's students and contribute to the high quality of instruction in the biology curriculum. Applicants must have a Ph.D. and a record of research accomplishments. Postdoctoral experience is preferred. Submit hard copies of a letter of application, statement of teaching philosophy, curriculum vitae, complete academic transcripts (student copy acceptable), representative reprints, and three letters of reference to: **Vertebrate Endocrinology Search, Dr. Patricia Edelmann, Chair, Department of Biological Sciences, California State University, Chico, Chico, CA 95929-0515.** Review will begin January 5, 2007. Electronic application will not be accepted. For full announcement see **website: http://csucareers.calstate.edu.** *For disability-related accommodations,* ***telephone: 530-898-6192 or TDD: 530-898-4666****. I-9/Equal Opportunity Employer/Affirmative Action/ADA.*

FACULTY POSITION
Insect Biodiversity

The Department of Entomology at North Carolina State University (NCSU) seeks to fill an **ASSISTANT PROFESSOR,** 12-month, tenure-track, research/teaching position in insect systematics. The incumbent will serve as Director of the NCSU Insect Collection. A Ph.D in entomology or related field and experience using diverse approaches in collections-based systematic research are required. Applicants must apply online. See **website: http://jobs.ncsu.edu** for instructions and required documentation. Applications will be accepted until January 2, 2007, or until a suitable candidate is selected. *Affirmative Action/Equal Opportunity Employer. ADA Accommodations: please call* ***telephone: 919-515-3148.*** *NC State welcomes all persons without regard to sexual orientation.*

POSITIONS OPEN

FACULTY POSITIONS
Environmental Genomics

As part of the Faculty Excellence Initiative at the University of South Carolina, applicants are being sought for three tenure-track positions at the **ASSISTANT PROFESSOR** level in the area of genome-environment interactions in aquatic systems. Interests include, but are not limited to, modern genomic approaches for assessment of impacts of pollutants, climate change, or other environmental stressors on humans or aquatic life, as well as research related to contaminant remediation strategies or bio/ecoinformatics. This is a joint search involving the School of the Environment and the Department of Biological Sciences in the College of Arts and Sciences and the Department of Environmental Health Sciences in the Arnold School of Public Health.

Applicants should submit a letter of application, curriculum vitae, statements of research and teaching interests, contact information for three references, and copies of selected publications to the: **Chair of the Environmental Genomics Search Committee, School of the Environment, University of South Carolina, Columbia, SC 29208,** or electronically to **e-mail: mgross@environ.sc.edu.** Review of applications will begin immediately and will continue until the positions are filled.

The University of South Carolina is an Affirmative Action, Equal Opportunity Employer. Minorities and women are encouraged to apply. The University of South Carolina does not discriminate in educational or employment opportunities or decisions for qualified persons on the basis of race, color, religion, sex, national origin, age, disability, sexual orientation, or veteran status.

MOLECULAR BIOLOGIST. Tenure-track **ASSISTANT PROFESSOR** position available August 2007. Ph.D. in molecular biology or related discipline; research interests related to bioinformatics or closely related area highly desirable. Successful applicant must be qualified to teach upper-division cell and molecular biology. Candidate is expected to establish an active, externally funded research program involving graduate and/or undergraduate students. Successful finalists must demonstrate effective communication and teaching by presenting a research seminar and a teaching demonstration during an on-campus interview. Mail one copy of all university transcripts, statements of teaching and research philosophies, curriculum vitae, and three letters of recommendation to: **Dr. Leslie Slusher, Department of Biology, West Chester University, West Chester, PA 19383** (no e-mail applications). Review of completed applications begins on January 2, 2007, and continues until position is filled. For more details and full ad see our webpage at **website: http://www.wcupa.edu/scripts/vacancies/v- list.asp,** call **telephone: 610-436-2751,** or **e-mail: lslusher@wcupa.edu.** *Affirmative Action/Equal Opportunity Employer. Women and minorities are strongly encouraged to apply.*

MARGARET AND HERMAN SOKOL CHAIR IN MEDICINAL CHEMISTRY
Montclair State University
College of Science and Mathematics
Department of Chemistry and Biochemistry

The College of Science and Mathematics at Montclair State University is pleased to invite applications for the Margaret and Herman Sokol Chair in Medicinal Chemistry from outstanding scientists whose research is focused on the chemistry of living systems in the areas of pharmaceutical or medicinal chemistry. Appointment to the Sokol Chair will be effective September 1, 2007. For further information and application requirements see **website: http://www.montclair.edu/hr/jobopport.htm** (VF38). Contact person: **Dr. Jeffrey Toney, Sokol Professor and Chairperson, Department of Chemistry and Biochemistry, Montclair State University, Montclair, NJ 07043.** *Women and minorities are strongly encouraged to apply. Montclair State University is an Equal Opportunity/Affirmative Action Employer.*

POSTDOCTORAL FELLOWSHIPS
Department of Molecular and Cellular Biology
Harvard University

The Department is actively seeking Postdoctoral Fellows in a wide variety of fields within molecular and cellular biology. Some appointments are funded through research grants awarded to faculty members and are ordinarily for one year, sometimes renewable; other appointments are possible through individual postdoctoral fellowships.

For information on these research opportunities, application instructions, and the list of faculty members with current openings, please visit the molecular and cellular **biology website: http://www.mcb.harvard.edu/Jobs/PostDocs.html.**

Harvard University is an Equal Opportunity Affirmative Action Employer.

FACULTY POSITION
Experimental High Energy Physics
University of Illinois, Urbana-Champaign

The Department of Physics invites applicants for a **FULL-TIME TENURED or TENURE-TRACK FACULTY** position in experimental high energy physics, beginning as early as August 2007. The University of Illinois, Urbana-Champaign Department of Physics has active programs in both experimental and theoretical high energy physics (see **website: http://web.hep.uiuc.edu/).** The successful candidate must have the ability to teach effectively at both the undergraduate and graduate levels and lead a vigorous and significant research program. Our existing experimental efforts include both collider physics (ATLAS, CDF, CLEO-c, ILC) and astrophysics (DES, LSST). A Ph.D. or equivalent is required. Salary is open and will be commensurate with qualifications. Submission will be via the web **(website: https://my.physics.uiuc.edu/join/).** Applicants will submit curriculum vitae, a list of publications, a brief description of their research and teaching interests and plans, as well as names of three references who can provide letters of recommendation. If you do not have internet access, please contact the **Physics Departmental Office, telephone: 217-333-3760,** to make other arrangements for submitting your application. For full consideration, application materials must be received by January 15, 2007. *The University of Illinois is an Affirmative Action/Equal Opportunity Employer. Minorities, women, and other designated class members are encouraged to apply.*

FACULTY POSITION
Experimental Atomic/Molecular and Optical Physics
University of Illinois, Urbana-Champaign

The Department of Physics invites applications for a full-time tenure-track faculty position, beginning as early as August 16, 2007, in the area of experimental atomic/molecular and optical (AMO) physics. The appointment is for the **ASSISTANT PROFESSOR** level; an appointment at a higher level will be considered for an exceptionally well-qualified candidate. A Ph.D., or equivalent, is required along with the ability to teach effectively at both undergraduate and graduate levels and to conduct a vigorous and significant research program. For full consideration for the coming academic year, completed applications must be received before January 20, 2007. Salary will be competitive and commensurate with qualifications. Applications may be submitted via the web **(website: https://my.physics.uiuc.edu/join/),** applicant will submit curriculum vitae, publication list, a summary of research interests and accomplishments, and the names and addresses of three references. If you do not have internet access, please submit applications to: **Kate Freeman, Physics Departmental Office, 1110 W. Green, Urbana, IL 61801, 217-333-3760, fax: 217- 244-4293, e-mail: katefree@uiuc.edu.** *The University of Illinois is an Affirmative Action/Equal Opportunity Employer.*

ANIMAL PHYSIOLOGIST

The Department of Biology at William Paterson University (WPUNJ) invites applications for a tenure-track position at the **ASSISTANT PROFESSOR** level. Ph.D. required. Postdoctoral research and teaching experience preferred. The successful candidate is expected to develop a research program that involves students. Teaching responsibilities will include undergraduate and graduate courses in anatomy and physiology and in area of specialization. Facilities of the Department include an established mouse laboratory with a full-time Technician, electron microscopy suites, and well-equipped molecular biology laboratories. The Department offers B.S. and M.S. degrees in both biology and biotechnology. Applicants should submit curriculum vitae, statements of research interests and teaching philosophy, names, addresses, and telephone numbers of three references to: **Dr. Eileen Gardner, Chairperson, Department of Biology, Science Hall, William Paterson University, 300 Pompton Road, Wayne, NJ 07470.** Review begins immediately and continues until the position is filled. *WPUNJ is an Affirmative Action/Equal Opportunity Institution; women and minorities are encouraged to apply.*

STAFF SCIENTIST
Cell/Molecular Biology

A Staff Scientist position is available for a **CELL/MOLECULAR BIOLOGIST** in the Department of Biological Sciences at Carnegie Mellon University. Candidates will participate in the development of novel agents for in-vivo molecular imaging utilizing MRI. An M.S. or Ph.D. degree is required, with a background in a broad range of recombinant DNA techniques; construction of viral vectors and/or transgenic technologies; quantitative gene expression detection methods; mammalian tissue culture; strong scientific problem solving skills; ability to communicate results in a clear manner verbally and in writing; record of scientific achievement as documented by peer-reviewed journal publications.

Interested candidates should send curriculum vitae and names of three references to: **Dr. Eric T. Ahrens, Department of Biological Sciences, Carnegie Mellon University, 4400 Fifth Avenue, Pittsburgh, PA 15213 U.S.A., e-mail: eta@andrew.cmu.edu.** *Carnegie Mellon is an Equal Opportunity/Affirmative Action Employer.*

MOLECULAR GENETICS. The Duke University Marine Laboratory seeks a **RESEARCH SCIENTIST** to manage a new Marine Conservation Molecular Facility in Beaufort, North Carolina. Responsibilities include facility set-up and maintenance, training and supervision of students and technicians in the laboratory, participation in collaborative projects with faculty, and establishment of strong ties to molecular facilities on the Durham campus of Duke University. We seek an energetic individual who shows promise of continuing initiative and who has excellent organizational and communication skills. Candidates should have a Ph.D. in molecular genetics; postdoctoral experience is beneficial but not required. To apply, please submit a letter of application, curriculum vitae, and names of three references to: **C.L. Van Dover, Duke University Marine Laboratory, 135 Duke Marine Lab Road, Beaufort, NC 28516; e-mail: c.vandover@duke.edu.** Review of completed applications will begin 15 December 2006. *Duke University is an Affirmative Action/ Equal Opportunity Employer.*

POSTDOCTORAL POSITION available for studies of oncogenic signaling mediated by AKT kinases or functional studies of tumor suppressors involved in mesothelioma. Experience with molecular genetic techniques and/or protein function is necessary. Please send curriculum vitae, statement of research interests, and the names and addresses of three references to: **Joseph R. Testa, Ph.D., Human Genetics Program, Fox Chase Cancer Center, 333 Cottman Avenue, Philadelphia, PA 19111. E-mail: joseph.testa@fccc.edu.** *Equal Opportunity Employer.*

POSITIONS OPEN

UNIVERSITY OF CALIFORNIA, LOS ANGELES

POSTDOCTORAL POSITIONS are available immediately in the Department of Anesthesiology at University of California, Los Angeles (UCLA) to study the biology of cardiac arrhythmias and heart failure. Strong collaborations exist within the Cardiovascular Research Laboratories and Division of Molecular Medicine at UCLA, a group of over 30 faculty with diverse research interests in cardiovascular biology. Successful candidates should have a Ph.D. or M.D. with demonstrated research experience in the following areas: electrophysiology, patch-clamp, isolated cell physiology and cell culture, microscopy, and biochemistry. Experience in small animal surgery and experimentation is desirable. Please send curriculum vitae, cover letter, and contact information (including e-mail) for three references to: **Dr. Aman Mahajan, Department of Anesthesiology, UCLA, e-mail: amahajan@mednet.ucla.edu.** *All qualified applicants are encouraged to apply including women and minorities. UCLA is an Equal Opportunity Employer.*

MOLECULAR CARDIOLOGIST. The Division of Cardiology at the Weill-Cornell University Medical Center seeks highly qualified applicants (M.D., Ph.D., M.D./Ph.D.) for tenure-track positions in basic and/or translational cardiovascular research at the **ASSISTANT or ASSOCIATE PROFESSOR** level. The Molecular Cardiology Program is a multidisciplinary team currently devoted to human genetics, cardiovascular development, vascular biology, genomics, electrophysiology, and gene therapy. Successful candidates should have a track record of outstanding achievement in basic investigation and will receive a generous startup package including newly renovated laboratory space. Please send curriculum vitae to: **Bruce B. Lerman, M.D., Chief, Division of Cardiology, Cornell University Medical Center, 525 East 68th Street, Starr 409, New York, NY 10021.**

THE STANFORD BIO-X FELLOW PROGRAM

The Stanford Bio-X Program is an interdisciplinary bio-related research program connected to biology and medicine. The Bio-X Fellow Program attracts young, highly talented researchers after their Ph.D. or first postdoctoral experience to start an independent, yet integrated research program with the potential of groundbreaking impact on biosciences. The Fellows are expected to creatively make use of the interdisciplinary resources of the Bio-X program and community. In this solicitation we are seeking a Fellow with research goals focusing on microbial systems.

For details of the posting please see **website: http://biox.stanford.edu/grant/pdf/fellow-program.pdf.**

Please send applications including curriculum vitae, plan research, and names of references to: **Professor Alfred M. Spormann (e-mail: spormann@stanford.edu).** Application deadline is December 3, 2006.

POSTDOCTORAL FELLOWSHIP in molecular and developmental neurobiology at the Center for Neuroscience Research, Children's National Medical Center, to study transcriptional regulation of oligodendrocyte development and myelination. The Center is a highly interactive scientific environment, with a strong program in developmental neurobiology. Candidates with experience in oligodendrocyte development and/or transcription factors in the nervous system are particularly encouraged to apply. For consideration, send curriculum vitae including list of publications, and the names/phone numbers/e-mail addresses of three people who could provide letters of reference to: **Vittorio Gallo, Ph.D., Center for Neuroscience Research, Children's National Medical Center, Room 5340, 111 Michigan Avenue N.W., Washington, DC 20010, fax: 202-884-4988, e-mail: vgallo@cnmcresearch.org.**

POSITIONS OPEN

ALBERT EINSTEIN COLLEGE OF MEDICINE
Advancing science, building careers

PSYCHIATRIC GENETICS
Research Fellowship

The Albert Einstein College of Medicine, Department of Psychiatry and Behavioral Sciences, Psychiatric Genetics Research Division is seeking a **FELLOW** (PGY-5 Level) for a one-year research Fellowship, beginning July 1, 2007. This Fellowship is available to M.D.s who will have completed their residency training by June 30, 2007. *Foreign graduates welcome but must be certified.* The Fellow will carry out research in the genetics of schizophrenia, bipolar disorder, and addiction. Some experience in molecular genetics research preferred but not essential.

Salary ranges from: $71,507 to $76,329.

Send or e-mail curriculum vitae, letter of interest, and three letters of recommendation to:

Dr. Herbert Lachman, Director
Laboratory of Behavioral Genetics
Albert Einstein College of Medicine
Department of Psychiatry and Behavioral Sciences
Jack and Pearl Resnick Campus
1300 Morris Park Avenue
Bronx, NY 10461
Fax: 718-430-8772
E-mail: lachman@aecom.yu.edu

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POSTDOCTURAL POSITION

The University of Maryland Center for Environmental Science (UMCES) Appalachian Laboratory (AL) invites applications for a Postdoctoral Position in watershed ecology. The successful candidate will join an interdisciplinary team to integrate biotic, hydrologic, and biogeochemical observations of the Potomac River watershed. The goal is to assess the impacts of land-use and land-cover change on this large river ecosystem. Experience working in interdisciplinary groups and with large data sets will be preferred. The position will be filled for one year with a possible second year renewal and salary will be competitive, commensurate with experience. Please submit an application electronically, with curriculum vitae, statement of research interests, and the names and contact information of three references, to **e-mail: orndorff@al.umces.edu, c/o Dr. Robert H. Gardner.** For further information visit our **website: http://www.al.umces.edu/employment.** Review of applications will begin immediately and continue through 1 March 2007, or until position is filled. *UMCES AL is committed to Affirmative Action, Equal Opportunity, and the diversity of its workforce.*

POSTDOCTORAL FELLOW
Wildlife Disease Laboratories at Conservation and Research for Endangered Species/ San Diego Zoo's Wild Animal Park
Position: 173034, Closes December 7, 2006, at 4 p.m.

We are seeking a Postdoctoral Fellow (Ph.D. or D.V.M./Ph.D.) to join the Molecular Diagnostics Laboratory. Projects center on discovery and characterization of etiologies and mechanisms of emerging and ongoing infectious and genetic diseases in diverse animal species. For additional info please visit our **website: http://www.sandiegozoo.org/employment.** Send curriculum vitae and names of three professional references to: **San Diego Zoo's Wild Animal Park, Attn: HR#173034, 15500 San Pasqual Valley Road, Escondido, CA 92027-7017, fax: 760-796-5614.** *Equal Opportunity Employer.*

POSTDOCTORAL POSITIONS available to investigate extracellular matrix components and inflammatory mediators in tissue injury and repair. Proficiency in cell and molecular biology is required. Please send a resume and three references to: **Bernadine Chmielowicz, Department Pharmacology and Toxicology, Rutgers University, 170 Frelinghuysen Road, Piscataway, NJ 08854** or **e-mail: bachmiel@eohsi.rutgers.edu.**

POSITIONS OPEN

POSTDOCTORAL FELLOW

(26UC3457) University of Cincinnati College of Medicine, Molecular Genetics. Research position to explore the function of vertebrate telomere proteins by making cell lines in conjunction with biochemical analysis. These studies will reveal the role of individual telomere proteins in telomere length regulation, telomere signaling, and telomere end protection.

Minimum qualifications: Ph.D. or M.D. with a strong background in biochemistry and molecular biology and must have experience in DNA cloning, PCR, and tissue culture. Good oral and written English language skills are essential.

For additional information and to apply for position number 26UC3457, please see **website: http://www.jobsatuc.com.** *Equal Opportunity Employer.*

POSTDOCTORAL RESEARCH ASSOCIATE
Cardiovascular Physiology

A Postdoctoral Research Associate position is available to study molecular basis of hypertension and heart diseases. Our research team stands in the forefront of hypertension research. This position has great potential for advancement and promotion based on accomplishments of first two years. Competitive salary and health insurance coverage for family. Ph.D. or/and M.D. Prior experience with at least two of the followings is preferred: gene therapy, stem cells, signal transduction, molecular biology, or cardiovascular physiology. Send detailed curriculum vitae and three references to: **Dr. Zhongjie Sun, Department of Physiology, P.O. Box 26901, BMSB 662A, University of Oklahoma Health Science Center, Oklahoma City, OK 73190 U.S.A. E-mail: zhongjie-sun@ouhsc.edu.**

POSTDOCTORAL FELLOWSHIP in the Section of Atherosclerosis, Department of Medicine, Baylor College of Medicine. Qualified applicants should have a Ph.D. or M.D. and experience in molecular biology with an interest in lipoproteins, inflammation, obesity, and vascular biology. Highly competitive salary will be offered and is negotiable depending upon experience. *Eligibility for NIH training-grant position with U.S. citizenship or residency required.* Reply with curriculum vitae and three references to: **Christie M. Ballantyne, M.D., Fellow of the American College of Cardiology, Professor, 6565 Fannin, M.S. A601, Houston, TX 77030. E-mail: cmb@bcm.tmc.edu.** *Baylor College of Medicine is an Equal Opportunity/Equal Access/Affirmative Action Employer.*

MARKETPLACE

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PrimeSTAR™ HS DNA Polymerase, a novel new high fidelity PCR enzyme which provides maximum fidelity as well as extended product length (8.5 kb for human genomic DNA; 22 kb for λ DNA). Targeted for demanding cloning (i.e. amplification of cDNA libraries) and sequencing applications, PrimeSTAR™ HS offers extremely high accuracy, excellent amplification efficiency and shortened reaction times.

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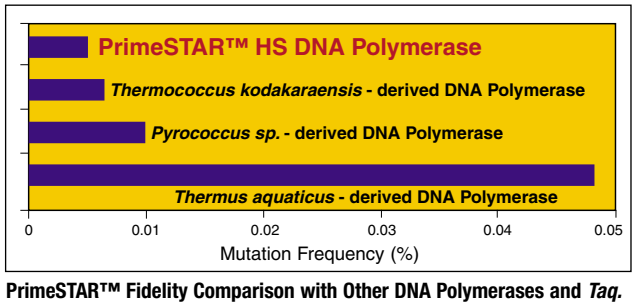
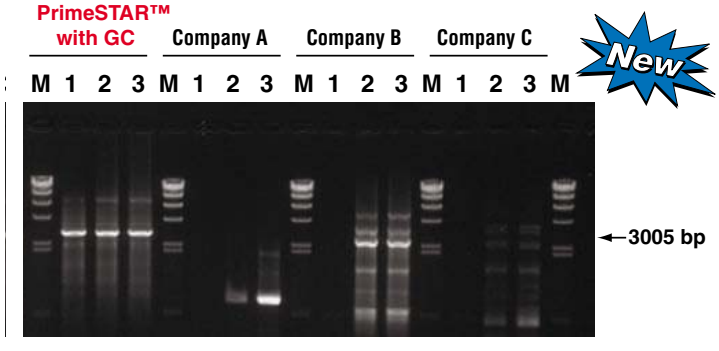
- **High Efficiency:** Higher than *Taq* Polymerase.
- **Robust Amplification:** A single PCR cycling protocol can be used to amplify products of varying sizes.
- **GC-Rich Targets:** Robust performance even with GC-rich templates.



PrimeSTAR™ Fidelity Comparison with Other DNA Polymerases and *Taq*.



Amplification of a 3005 bp high-GC (73.2%) TthHB8 genomic DNA template. The performance of high fidelity, high-GC enzymes from Companies A, B, and C was compared with PrimeSTAR™ HS DNA Polymerase with GC Buffer. Lanes 1, 2, and 3: 100 pg, 1 ng, 10 ng human genomic DNA template.

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